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The effects of nitrogen and DDT on the development rate and fecundity of tetranychus telarius (linaeus) on chrysanthemums.

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THE EFFECTS OF NITROGEN AND DDT ON THE DEVELOPMENTAL RATE AND FECUNDITY OF TETRANYCHUS TELARIUS (LINNAEUS) ON CHRYSANTHEMUMS

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THE EFFECTS OF NITROGEN AND DDT ON THE DEVELOPMENTAL RATE AND FECUNDITY OF *TETRANYCHUS TELARIUS* (LINNAEUS) ON CHRYSANTHEMUMS

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INTRODUCTION

The complex nature of population dynamics encompasses many diverse interactions between a species and its environment. The factors involved in the close relationship between phytophagous arthropods and their host plants are not well defined.

Recent investigations have indicated that phytophagous mite populations are strongly influenced by the nutritional status of the host. Mite fecundity and population responses have been closely linked to the amount of nitrogenous materials available to the plant. It has been suggested that pesticides may affect plant physiology and that low concentrations of DDT may stimulate plant growth in a manner not unlike that which occurs as a result of fertilization with nitrogenous substances.

Many workers have felt that resurgent mite populations are due not only to the elimination of natural enemies and interspecific competition, but also to some unknown physiological action of the pesticide upon the plant. The possible interrelationship between DDT, nitrogen, and mite population fluctuations has received relatively little attention.

With these factors in mind, it was the purpose of the study reported herein to investigate further the effect of

nitrogen upon the nutritional status of the host plant in terms of the developmental rate and fecundity of mites. Additional investigations were undertaken to determine the effects of DDT on the nitrogen content of the foliage and the resulting response of the mites thereon.

Chrysanthemums were grown in sand, soil, and vermiculite cultures at several levels of nitrogen and with various DDT treatments. The response of the two-spotted spider mite, Tetranychus telarius (Linnaeus), to the treatments was ascertained on both detached and intact foliage, and correlations were made between the nitrogen content of the foliage and the mite response.

REVIEW OF THE LITERATURE

The relationship between phytophagous mites and their host plants has received increased attention during recent years. This interest may be attributed, in part, to the need for a more complete understanding of the resurgent and resistant populations which occasionally develop following pesticide applications. Such problems have led entomologists to intensify the study of the basic factors underlying the dynamics of arthropod population fluctuations. The following sections are devoted to a review of the literature concerning the relation between mite populations and natural enemies, host plants, nitrogen, and DDT.

NATURAL ENEMIES.

The major triggering mechanism for resurgence appears to be the sometimes catastrophic effect of pesticide applications upon the natural enemies of the resurgent populations. The importance of natural enemies in regulating pest abundance and the lethal effect of DDT (dichlorodiphenyl-trichloroethane) on many beneficial arthropods is well established (Clancy and Pollard, 1948; Lord, 1949; DeBach *et al.*, 1950; Pickett and Patterson, 1953; Collyer, 1953, 1958; Collyer and Kirby, 1959; and Weidhaas, 1959).

The literature concerning this phase of the problem has been reviewed by MacPhee and Sanford (1954), Ripper (1956), and Weidhaas (1959). The latter two workers have tabulated the reports of resurgent mite populations.

HOST PLANT.

It has been demonstrated that an important factor in regulation of phytophagous arthropod populations is the nutritional status of the host plant. Significant correlations between plant nutrition and insect fecundity, developmental rate, or attack have been established (Haseman, 1946; Dahms, 1947; Arant and Jones, 1951; Barker and Tauber, 1951; Mittler, 1953; Daniels and Porter, 1956; Auclair et al., 1957; and others). Rodriguez (1960) has recently reviewed the literature concerning the nutrition of the host plant and reaction to pests. The known information concerning insect and mite nutrition has been reviewed rather completely (Uvarov, 1928; Craig and Hoskins, 1940; Wigglesworth, 1950; Trager, 1953; Lipke and Fraenkel, 1956; Friend, 1958; Gordon, 1961; and House, 1961).

Species. Whitcomb et al. (1940, 1941, 1944) noted that both the developmental rate and the fecundity of T. telarius varied with the species of host plant. Roses afforded mites faster development and greater fecundity

than gardenias or snapdragons; mites produced twice as many eggs on beans as on lettuce or cucumbers. Similar observations on roses were made by Neiswander et al. (1950). Fritzsche et al. (1957) found that the degree of mite infestation depended on the variety of bean fed upon, and Kuenen (1949) reported a similar relationship between apple varieties and the European red mite, Panonychus ulmi (Koch).

Whitcomb et al. (1940) and Neiswander et al. (1950) reported that T. telarius was more difficult to kill with pesticides on roses than on other plants. Saba (in Gordon, 1961, p. 49) found that T. telarius after twelve generations developed high resistance (50-200x) on nettle and hop and low resistance (5-10x) on bean, carnation, and European plum to tetraethylpyrophosphate (TEPP).

pH. Whitcomb et al. (1943) found that the rate of development was correlated with the pH of the plant sap and that T. telarius developed faster on low than on high pH sap on both roses and carnations. Further investigation, however, indicated that this correlation was not consistent among greenhouse plants and suggested that additional factors were involved (Whitcomb et al., 1944).

Leaf age. Leaf age has been shown to exert some influence upon the fecundity and developmental rate of mites. Young citrus leaves were found to be more suitable for the

citrus red mite, Panonychus citri (McGregor), than were mature or damaged (bronzed) leaves on which less eggs were produced (Henderson and Holloway, 1942; Fleschner, 1952). Jeppson et al. (1953) also reported young citrus leaves to be more favorable for the development of P. citri than mature leaves, and added that population trends were correlated with the periods of maximum new growth (Jeppson et al., 1957). P. ulmi was reported by Miller (1953) to have developed more rapidly on undamaged foliage than on bronzed foliage. Bronzed or senescent foliage triggered the onset of diapause with P. ulmi (Lees, 1953).

Plant vigor. Plant "vigor" has been cited as a factor which influences mite abundance. Kuenen (1949) reported that P. ulmi egg production was much greater on healthy vigorous trees than on neglected trees, and Roesler (1953) stated that pruning and cultivation of fruit trees encourages mites by increasing the amount of sunlight and warmth in the crowns. Reed (1936), however, observed that P. ulmi was three times more abundant on moderately vigorous trees than on vigorous trees, and indicated that good culture to increase the vigor of the tree was an important element in mite control.

NITROGEN

The approach to an understanding of population fluctu-

ations by alteration of the plant substrate is not new, for McGregor (1914) had attempted to control mites by fertilizing the soil. Although McGregor's results were inconclusive, Hoffman (1918) reported that manurial applications to fruit trees encouraged red spider (Tetranychus spp.) infestation. Speyer (1926), on the other hand, found that T. telarius infestations on tomatoes were not affected by manurial treatments.

According to Rodriguez (1960, p. 156) Bondarenko in 1949 indicated that heavy mineral fertilization can increase the osmotic pressure of plant sap and also increase populations of T. telarius. A systematic survey of Ohio greenhouses by Rodriguez and Neiswander (1949) indicated that mite populations were correlated with the soluble salt content of the soil, and a subsequent reduction in the amount of fertilization was accompanied by reduced populations.

Tomato. In controlled sand culture studies with T. telarius on tomatoes Rodriguez (1951) found that while mite fecundity showed no significant differences at the 100, 200, and 400 ppm. levels of nitrogen, an increase to 800 ppm. caused a severe reduction in reproductive capacity. Further studies with tomato in sand culture showed that while increased nitrogen supply was positively correlated

with vitamin elaboration (niacin and riboflavin) the relationship of these vitamins to T. telarius populations was inconclusive (Rodriguez and Rodriguez, 1952).

Bean. Garman and Kennedy (1949) found that fertilization of beans in soil culture with 7-5-9 (N:P:K) fertilizer resulted in a slight increase in T. telarius populations after two weeks and a three-fold increase after three weeks when compared to the unfertilized controls. Studies in sand culture yielded similar results. Further work with T. telarius (Garman, 1959) showed that plants in sand culture produced fewer mites when fed with organic nitrogen compounds (asparagine and monosodium glutamate) than those which received calcium nitrate. Fritzsche et al. (1957) reported that T. telarius populations on beans in sand culture were positively correlated with the nitrogen content of the foliage (also with the content of reducing sugars). In addition, field experiments with two varieties of beans yielded greater mite populations on the variety which contained more nitrogen (and more reducing sugars) in the foliage. Henneberry and Smith (1960) reported that the reproductive rate of T. telarius reared on beans in sand culture increased as the nitrogen supply to and the absorption by the plant increased (the reproductive rate was also positively correlated with increased concentrations of water soluble carbohydrates

in the foliage).

Henneberry and Stuart (Anon., 1958) found that pole bean plants grown at nitrogen levels sufficient for normal growth produced T. telarius populations which were easier to kill with malathion than those from plants supplied with excessive or deficient levels of nitrogen.

Apple. Hamstead and Gould (1957) applied various levels of nitrogenous fertilizer to apple trees in the orchard and found that the resulting populations of T. telarius and P. ulmi were positively correlated with the nitrogen content of the foliage, but the developmental rate was not affected. In addition, they noted that the seasonal population peak for P. ulmi coincided with that for nitrogen content of the foliage; T. telarius populations peaked two weeks later. Rodriguez (1958), using discs of leaves detached from apple plants grown in sand culture, found that increased nitrogen supply resulted in increases in T. telarius populations and foliar nitrogen. He noted that the response of T. telarius to the variation in nitrogen was much more pronounced and sensitive than that of P. ulmi. However, Hukusima (1958) found that while apple plants growing in water culture produced higher populations of arthropods at the higher levels of nitrogen, the population of T. telarius was greater on nitrogen-deficient plants; P. ulmi, on the other hand, produced more

offspring as the nitrogen level increased. Utilizing both intact foliage and discs from detached apple leaves, Breukel and Post (1959) found that the increased nitrogen content of the foliage produced on manure-fertilized plots resulted in higher egg production and decreased egg, larval, and nymphal mortality of P. ulmi. The higher nitrogen level also increased the rate of development but not significantly.

Peach. Peaches grown in sand culture produced larger populations of T. telarius with heavier fertilizer treatments than with light fertilization (Garman and Kennedy, 1949).

Cucumber. LeRoux (1954) found that increased fecundity of T. telarius was significantly correlated with increased nitrogen supply to cucumbers grown in sand culture.

Chrysanthemum. No studies have been reported concerning mite populations and the nitrogen nutrition of chrysantheumums.

DDT

Direct effect on mites. The reports concerning direct mite response to DDT-treated, predator-free plants are inconsistent. Huffaker (1948) found no increase in the fecundity of T. telarius when it fed upon foliage

which had DDT in its translocation stream. Direct spraying of soybeans severely reduced the population of T. telarius (Wingo and Thomas, 1948), and Boudreaux (1956) reported little or no effect upon longevity, oviposition rate, total volume of eggs, or hatching when spider mites were treated with DDT. However, Hueck (1953) reported that on apple trees, from all of which predators had been eliminated, the first generation of P. ulmi which developed following a 0.1 per cent DDT spray laid more eggs than on non-DDT trees. Similar findings occurred in the laboratory at low concentrations of DDT, but high concentrations caused population reductions.

Davis (1952b) found that DDT irritated T. telarius and caused a temporary reduction in oviposition. This irritation resulted in a scattering of the colony and, because this mite produces more eggs in non-crowded conditions (Davis, 1952a), the overall effect of the DDT application was a threefold increase in the population as compared to the untreated colonies.

Effect on plants and indirect effect on mites. Since the initiation of its use, DDT has been repeatedly connected with various degrees of phytotoxicity. The sensitivity of cucurbits to DDT has been well established and many other crops have been injured by foliar or soil applications. The accumulated literature concerning DDT

phytotoxicity has been reviewed by many investigators (Carruth and Hervey, 1947; Allen et al., 1951; Foster, 1951; Weigel et al., 1951; Allen et al., 1954; Ginsburg, 1955; MacCollum, 1956; and others). While it may be injurious at normal or excessive dosages, DDT appears to act like a growth stimulant at low concentrations.

Wheeler (1945) reported that DDT appeared to cause either a stimulation of vegetative growth or retardation of the ripening or maturing processes when used on quince. Similar effects were noted on onion (Wilcox and Howland, 1950), squash (Walton, 1947), squash and muskmelon (Bruce and Tauber, 1945), tobacco (Young and Gill, 1948), and other vegetables (Allen, 1947; Krishnaswami, 1954). Root stimulation of cucumber seedlings in petri dishes was reported by Casida and Allen (1951) and Sazonov et al. (1956) reported similar findings with wheat seed germination. Plant stimulation occurred at different, but definite, levels of DDT for each plant tested by Chapman and Allen (1948); in addition, they noted that DDT appeared to have been taken into the plant through the roots and caused stimulation of the aerial portions of the plant when the proper dosage was used.

The presence of DDT in root crops and in the foliage of crops grown in treated soils indicates the DDT may be translocated within plants. Casida and Allen (1952) stated

that accumulated evidence indicated that the causative agent for plant stimulation by DDT must be absorbed and translocated. While there are many negative reports concerning the translocation of DDT (Hoskins, 1949; Weigel et al., 1951; and others), absorption into various crops has been reported (Burt and Ward, 1955; Lichtenstein, 1959; Satyanarayana, 1954). Muns et al. (1960) detected DDT in the foliage of sugar beets grown in DDT-treated soil. Kozlova and Dvortzova (1952) reported systemic activity of DDT in corn with the subsequent death of the green peach aphid, Myzus persicae (Sulzer), feeding on the foliage. Huffaker (1948) was able to cause DDT translocation in beans by artificial methods. The mechanics involved in the process of DDT absorption and translocation are not completely understood, but the evidence available indicates that such phenomena do occur. Thus, the possibility of a physiological response of plants which might favor mite increases appears plausible.

An indirect effect through changes in the physiology of the host plant has been proposed to account for mite resurgence following DDT applications. Several workers have reported that the lack of predation was not sufficient to explain mite increases and that physiological changes within the host plants were probably involved (Huffaker and Spitzer, 1950; Fleschner, 1952; Michelbacher et al., 1952;

Klostermeyer and Rasmussen, 1953; and Tew and Groves, 1956).

Nutritional sprays were found to trigger P. citri increases (Thompson, 1939; Holloway et al., 1942). Fleschner (1952) felt that the deposits of field dusts, DDT, and nutritional sprays, which were accompanied by P. citri increases, lowered the resistance of citrus trees owing to some physical properties of the materials. Furthermore, he noted that the lowered resistance of the older foliage spread to the younger leaves. Warren and King (1959) found that mechanical factors such as pubescence or surface residues significantly influenced the development of Tetranychus hicoriae (McGregor) infestations, but that there was no physiological response of the pecan tree involved.

DDT-NITROGEN-MITE COMPLEX.

The reports cited above indicate that: (1) nitrogen is an important factor in mite reproductive capacity; (2) DDT can stimulate plant growth in a manner not unlike stimulation by nitrogen; and (3), DDT may cause physiological changes in the foliage which enhance the biotic potential of mites. In addition, MacCollum (1956) reported that DDT-injured cucumber foliage had higher nitrate

concentrations than normal foliage. These findings suggest that DDT may cause changes in the nitrogen content of the foliage which enhance the biotic potential of mites.

Investigation of a possible DDT-nitrogen-mite complex was initiated by Rodriguez et al. (1957). These workers found that DDT tended to depress nitrogen in the foliage of soybeans (at 80 - 100 pounds/acre) and black valentine beans (at 5 - 100 pounds/acre) and increased nitrogen in bean foliage at higher levels. On detached leaf discs T. telarius tended to be negatively correlated with the nitrogen content of the bean foliage. Apple trees grown in soil treated with heavy applications of DDT (100 and 4000 pounds/acre) had more nitrogen in the foliage than did trees grown in non-treated soil (Rodriguez et al., 1960a). Detached leaf discs from the treated trees produced higher T. telarius and P. ulmi populations than did discs from untreated trees. Other studies (Rodriguez et al., 1960b) showed that 800 pounds of DDT/acre increased the nitrogen content of bean foliage (also the total and reducing sugar content) and increased T. telarius populations on leaf discs.

METHODS AND PROCEDURES OF INVESTIGATION

GENERAL

The objectives of this study were to determine the effects of: (1) nitrogen on the host plant, chrysanthemum; (2) host plant nutrition on mite fecundity and developmental rate; and, (3) DDT on host plant nitrogen content and on mite fecundity and developmental rate.

To determine the host plant response to nitrogen, chrysanthemums were grown in vermiculite and fed with known nutrient solutions. The response of the plants to the various nutrient solutions in terms of growth and foliar content was determined.

A second test was devised to study the effect of host plant nutrition on the mite. Plants were grown in sand culture and fed with nutrient solutions. Mites were then placed on detached leaves under controlled conditions for observation.

The combined effect of nitrogen and DDT was investigated by first treating the sand substrate with DDT and then feeding the plants with nutrient solutions. The foliage was detached and mites were introduced under controlled conditions for observation.

In order to check the reliability of the detached leaf method, a greenhouse test was conducted. Mites were

placed on isolated leaves of the host plants under the various nutrient and DDT treatments.

Finally, a test was designed to indicate the practical significance of the findings of the previous tests. Chrysanthemums were grown under conditions which approached normal commercial situations. Liquid nitrogenous fertilizer was applied to the soil at recommended and at surplus rates. These plants were both naturally and artificially infested and the mite response was ascertained.

For the sake of clarity in the ensuing discussion the five tests described above will be referred to as the Vermiculite series, the Nitrogen series, the Nitrogen x DDT (detached) series, the Nitrogen x DDT (intact) series, and the Liquid Nitrogen series, respectively.

PLANT CULTURE

General.

Cuttings of Chrysanthemum morifolium variety Illini Cascade were secured from Yoder Brothers, Inc., Barberton, Ohio, for these studies. The original cuttings served as test plants and stock plants for the vermiculite and sand culture experiments. New cuttings were ordered from the same source for the Liquid Nitrogen series.

In vermiculite and sand culture, the plants were

grown in one-gallon glazed crocks which had a drainage hole on the side at the base. Water, de-ionized by passage through the resinous filter of a Barnstead Bantam De-ionizer, was used with the vermiculite and sand cultures for nutrient solutions and daily watering. Tap water was used in the Liquid Nitrogen series.

Nutrient solutions (adapted from: Hill et al., 1934; Stuart, 1948; Hoagland and Arnon, 1950; and Asen and Wildon, 1953) were prepared at weekly or bi-weekly intervals from 0.5 M stock solutions which were stored in aluminum-painted glass bottles in a shaded part of the greenhouse. A complete minor element solution (Hoagland and Arnon, 1950) and chelated iron were added to all nutrient solutions for culture in artificial substrates. Ammonium nitrate and potassium chloride stock solutions in the Liquid Nitrogen series were stored in carboys under a black cloth in the greenhouse. In all series the nutrients were applied in measured dosages directly onto the substrate and water was added as needed.

Overhead lamps with reflectors provided three to four hours of supplementary light nightly to encourage continuous vegetative growth (Andrease et al., 1956). Except for the plants of the Liquid Nitrogen series, all plants were "soft-pinched" to encourage the production of three or more stems.

Temperature and humidity records were obtained in the laboratory and greenhouse with a Bendix-Friez hygrothermograph model 594.

Vermiculite series.

For this study vermiculite (Terralite, Grade 2) was the substrate. Three cuttings were placed in each of twenty crocks which were then placed at random on boards which rested on the greenhouse bed, approximately one foot above ground level. Rooted cuttings were potted in the vermiculite on October 7, 1958 and continued for sixteen to eighteen weeks.

Nutrient solutions were added to the substrate weekly at the rate of 250 ml. per crock. The following nitrogen levels were utilized: 25, 50, 100, 200, and 400 ppm. These nutrient solutions, hereafter referred to as Method A, contained the millimolar concentrations of nutrients which are indicated in Table 1.

Table 1. Number of millimoles per liter of nutrient solution. Method A.

Nutrient	Nitrogen Level (ppm.)				
	25	50	100	200	400
KNO_3	0.90	1.79	3.58	7.15	14.30
KH_2PO_4	1.00	1.00	1.00	1.00	1.00
$Ca(NO_3)_2$	0.44	0.90	1.79	3.58	7.15
$MgSO_4$	2.00	2.00	2.00	2.00	2.00
KCl	13.40	12.52	10.72	7.12	0.00
$CaCl_2$	6.70	6.26	5.36	3.58	0.00

By formulating the nutrient solutions with changes in the calcium and potassium salts, the increase in nitrogen was accompanied by a decrease in chlorine. Thus, at 25, 50, 100, 200, and 400 ppm. levels of nitrogen there were 951, 888, 761, 507 ppm., and a trace of chlorine, respectively. Other major nutrients were present in the following amounts (ppm.): K, 598; P, 31; Mg, 49; Ca, 287; S, 48. The pH of the nutrient solutions ranged between 5.0 and 5.7.

Sand culture. (Nitrogen and Nitrogen x DDT (detached and intact) series.)

The substrate in these studies was a 1:1 mixture of #1 and #3 quartz sand. Each crock contained two plants and was supported on raised branches by an inverted clay pot (Fig. 1a), both surfaces of which received heavy bands of Tanglefoot to prevent plant contamination by stray mites. In addition, a heavy band of Tanglefoot was applied to the exterior of each crock. When possible, the crocks were placed in randomized blocks to minimize the effects of any environmental factors within the greenhouse itself. Paraffin-coated dowels, $\frac{1}{4}$" x 3', were inserted into the substrate to act as supports to which the plant could be tied.

Nutrient solutions were provided using Method A, as described in the previous section, or Method B, in which ammonium nitrate furnished nitrogen increases. One group

a.

b.

FIGURE 1

a. Experimental arrangement for Nitrogen x DDT (intact) series.

b. Experimental arrangement for Liquid Nitrogen series.

of plants received Method A solutions in the Nitrogen series, while the other group received Method B solutions. Only Method B was used in the two Nitrogen x DDT series. The conversion to Method B for the bulk of the experimental work eliminated the changes in chlorine content in the nutrient solution which existed with Method A, with the result that nitrogen was the only nutrient which varied. Table 2 indicates the millimolar concentrations of the nutrients under Method B.

Table 2. Number of millimoles per liter of nutrient solution. Method B.

Nutrient	Nitrogen level (ppm.)				
	25	50	100	200	400
NH_4NO_3	0.00	0.89	2.68	6.24	13.40
$NH_4H_2PO_4$	0.89	0.89	0.89	0.89	0.89
KNO_3	0.89	0.89	0.89	0.89	0.89
$MgSO_4$	2.00	2.00	2.00	2.00	2.00
KCl	5.00	5.00	5.00	5.00	5.00
$CaCl_2$	3.50	3.50	3.50	3.50	3.50

One half of the nitrogen concentration at each level was nitrate-nitrogen and one half ammonium-nitrogen. Major nutrients, other than nitrogen, were present in the following amounts (ppm.): K, 240; P, 28; Mg, 49; Ca, 140; S, 48; Cl, 425. The pH of the nutrient solutions ranged from 5.2 to 5.9.

The plants grown under Method A were planted in October, 1958 and were one year old at the time of these tests. These plants had been cut back to the original stem and allowed to branch out again. Those grown without DDT under Method B were planted in June, 1959, and those with DDT were planted in September, 1959.

The nutrients were applied at regular intervals twice a week at the rate of 250 ml. per crock per week. In March, 1960, a nutrient deficiency developed; both symptoms and leachate samples indicated that the deficiency was due to calcium. The plants had recently been relocated and improved conditions for plant growth at the new location indicated that the deficiency was brought about by the increased growth rate of the plants rather than an inherent imbalance of the nutrient solution. For this reason, the nutrient solutions were not altered, but the rate of application was increased to 143 ml. per crock three times weekly. With these adjustments the deficiency symptoms disappeared.

The crocks were leached at two to three week intervals in order to prevent the build-up of unused nutrients in the sand. Both the leachate from the crocks and the nutrient solutions were checked periodically for pH with a portable Beckman pH meter (model 180).

It was necessary to control insects which invaded the greenhouse and either infested the experimental plots or

were potentially dangerous to the experimentation. Under these conditions, the entire greenhouse was fumigated with calcium cyanide or Plantfume 103 (tetraethyl dithiopyrophosphate). A heavy band of Tanglefoot was applied to the outside of all crocks and to both the inside and outside of the supporting clay pot to prevent access by stray mites. In the Nitrogen x DDT (intact) series, a single barrier of plastic sheeting was used to prevent possible contamination by mites drifting from an adjacent rose bed.

Soil culture. (Liquid Nitrogen series.)

A split plot design was utilized for this experiment; the plots consisted of three nutrient treatments, and the subplots consisted of DDT treatments. Each subplot consisted of two rows of four plants each. Each plant was separated from its opposite by a distance of five inches, and from its adjacent member along the row by five and one-half inches. Each row was set ten inches from the row in the adjacent subplot. On either side of the subplot was a composition separator which extended into the soil approximately five inches. The separators acted as retainers for the nutrients which were applied to the subplot. A total of nine subplots made up one complete replicate; the experiment consisted of four such replicates, two on each of two twenty-five foot raised beds (Fig. 1b).

Within each replicate the plots were distributed randomly; the subplots were distributed randomly within each plot.

At either end of each bed was a check, which received neither nitrogen nor DDT treatments. Thus, although not a part of the split plot design, a total of four check plots was utilized.

A steam-sterilized Merrimac gravelly sandy loam to which sand and peat had been added (3 parts soil : 1 part sand : 1 part peat) was the substrate for these tests. Superphosphate and dolomitic limestone were mixed into the substrate at the rate of ten and five pounds per 100 square feet, respectively. Potassium chloride was applied at bi-weekly intervals to all plots at the rate of one and one-half pounds per 100 gallons of water, delivered at the rate of one quart per square foot.

Four levels of ammonium nitrate (0, 50, 200, and 400 ppm. N) were applied twice weekly at the rate of one quart per square foot of surface per application. The stock solutions consisted of 0, 38, 151, and 302 grams of ammonium nitrate, diluted to 15 quarts of aqueous solution at the 0, 50, 200, and 400 ppm. levels, respectively. Additional water was applied daily in the morning to the soil and in the afternoon as a light spray on the foliage.

A standard hose with a "Hozon" attachment was used to apply the ammonium nitrate and potassium chloride. This

attachment is a copper fitting with a side-tube which, when coupled between the hose and the water tap, acts as a siphon when the main water flow is turned on (Fig. 2). Thus, with the water flow on and the tubing immersed in the stock solution, the latter is drawn up and proportioned into the main flow at a rate of 1:17. Nutrient applications were made between 6 and 7 A.M. when there was a minimum of water pressure fluctuation, thus allowing the delivery to be timed rather than volumetrically measured. The time necessary to deliver the desired volume was determined anew prior to each day's application.

Rooted chrysanthemum cuttings were planted on June 15, 1960. Nutrient applications were initiated on July 1, and continued until after the plots had been sampled and testing had been completed in September. These plants were grown on a single stem, with new side breaks being removed as they formed. Each plant was supported by a string connected at the base of the plant and tied to an overhead wire which ran the length of the bench. Petrolatum was applied along the wires at points between each plot in order to restrict mite movement.

With the exception of stock chrysanthemums, no other plants were grown in this compartment of the greenhouse during the experimental period.

Insect control of these plots was necessary as aphid

a.

b.

FIGURE 2

a. Hozon proportioner. Close-up.

b. Hozon proportioner. In operation.

and cutworm infestations occurred. The cutworms, Prodenia ornithogalli Guenée, were removed by hand. This primitive method was quite rewarding as this species did not return to the soil during the day and thus was easily located. The aphids, Macrosiphoniella sanborni (Gillette), Anuraphis helichrysi (Kaltenbach), and Aphis gossypii (Glover) were not easily discouraged, however, and it became necessary to fumigate the greenhouse with Nicofume on August 3, 1960. This treatment had no discernible effect on the test mites or their insect predators.

DDT APPLICATIONS

Sand culture. (Nitrogen x DDT (detached and intact) series.)

DDT was applied to the sand at the rate of 0.9, 5, 10, 20, and 40 pounds per acre on September 20, 1959. Rooted chrysanthemum cuttings were planted the same week. A single crock containing two plants was used to test the effects of the insecticide at each of four nitrogen treatment levels (50, 100, 200, and 400 ppm.). Thus, for each DDT treatment a total of four crocks was prepared.

The exposed surface area of the sand in four crocks was 3.97 square feet. DDT solutions for each four crock unit were prepared as indicated in Table 3. These solutions were prepared by first dissolving the DDT in acetone

and then adding water as needed.

Table 3. Amounts of technical DDT, acetone, and water applied to each four crock batch of sand.

DDT level lbs./A.	DDT grams	Acetone (plus DDT) ml.	Water ml.
0.9	0.0372	150	100
5.0	0.2068	150	100
10.0	0.4137	150	100
20.0	0.8274	225	100
40.0	1.6547	250	0

The DDT solution was applied with a DeVilbiss atomizer to small batches of the sand from four crocks until all the sand had been treated and all the solution had been used. When all the sand of one DDT level had been treated, it was thoroughly mixed in a revolving twenty-gallon can. The treated sand was then returned to the four crocks which would receive the four different nutrient solutions. The application equipment was thoroughly cleaned with acetone between DDT treatments.

Soil culture. (Liquid Nitrogen series.)

Sub-plot treatments consisted of the application of DDT to the soil, a DDT foliar spray, and a control which received no DDT. Each DDT treatment was replicated four times at each nutrient level. The check plots at the ends

of the benches, which received no additional nutrients, were not treated with DDT.

DDT was applied to the soil at the rate of 20 pounds of active material per acre. Each treated plot (3.33 square feet) received 1.392 grams of 50 per cent wettable powder. The application was made after the soil had been prepared and sterilized but before the introduction of the rooted cuttings. The DDT for a single plot was thoroughly mixed with approximately one pint of prepared soil which served as a diluent, and then evenly spread over the entire surface of the plot. The freshly prepared soil was then turned by hand for several minutes to mix the DDT thoroughly and evenly throughout the entire plot. The application was made on June 15, 1960, and the rooted cuttings were planted later on the same day.

The subplots which were to receive DDT as a foliar application were sprayed with a 50 per cent wettable powder at the rate of one pound of actual DDT per 100 gallons of spray on August 5. Triton B-1956 was added as a wetting agent. The spray material was applied to the point of run-off with a hand operated, one and one-half gallon capacity, compressed air sprayer. Both the upper and lower leaf surfaces received complete coverage. A plastic cage was used to confine the spray to the plot being treated; a

loose, but overlapping, flap at one end of the cage allowed the operator freedom of movement with the equipment while at the same time restricting the drift of the spray material to the confines of the plot. Approximately 0.1 quart of spray material was applied to each plant sprayed.

MITE INTRODUCTION AND EXPERIMENTAL CONDITIONS

General.

The two-spotted spider mite, Tetranychus telarius (Linnaeus), was the test animal for all experimental work. Several specimens of mites were taken from each experimental series and were subsequently identified by the author as T. telarius. These determinations were verified by Dr. E. W. Baker, U.S. National Museum, Washington, D.C.

The original mites for the culture were taken from chrysanthemums in a local commercial greenhouse. They were reared for several months on baby lima bean plants and then transferred to chrysanthemums. The mites used in the experimental work were all taken directly from chrysanthemums.

Plants for mite-rearing were propagated in both soil and vermiculite at a night temperature of 65°C. with three to four hours of supplementary light. These plants were located within a large cheesecloth cage to prevent in-

vasion by predatory and/or other insects. Furthermore, each pot was protected from contamination by stray mites by the use of water-filled moats within the cheesecloth enclosure.

When needed for experimental work, a surplus of male and female mites was transferred with a camel's hair brush from the rearing plants onto detached foliage in a petri dish. It was then possible to select the most active forms for experimental use and also to determine more accurately those which were approximately the same age with the aid of a stereoscopic microscope.

Mites selected for experimental work consisted of three young adult females and one male mite per introduction in the sand culture studies and two young adult females in the Liquid Nitrogen series.

Detached leaf procedure. (Nitrogen and Nitrogen x DDT (detached) series.)

Leaves were detached from plants in the greenhouse and placed directly on filter paper supported by approximately one-half inch of quartz sand in a petri dish. The sand was kept wet with de-ionized water. A restraining band of petrolatum was applied to the periphery of the filter paper.

Young leaves were utilized in all cases, i.e., the

first or second fully-formed leaf below the apical meristem of the plant. In this way it was hoped to minimize any undue error which might stem from variation in the age of the foliage. The leaves were detached late in the afternoon in order to minimize variation in the nutritional status of the foliage due to time of detachment. Having been detached, the leaves were then placed on the moist filter paper where they were checked for the presence of insects or other arthropods, which were removed if found. On the evening of the same day the adult mites were introduced individually as previously described.

After the petiole had been removed, the blade was cut into two sections, one larger than the other (Fig. 3). Test mites were placed on the larger portion, while the smaller portion was reserved for the adult progeny of the introduced mites. Thus, an adult female which was the daughter of the originally introduced mites was transferred to the small reserved portion to provide a new observation area for oviposition records. In this manner the original progenies were separated from those of the succeeding generation. This system made it possible to record accurately not only the developmental periods of the original progeny, but also the first appearance of second generation eggs and larvae.

The petri dishes, without covers, were then placed

FIGURE 3

Detached leaf sections in petri dish.

on a tray in a constant temperature cabinet (Fig. 4).

On the eighth day the original mites were removed, a population count was made, and the developmental stages of the mites present were recorded. Subsequently, daily records were made of the stages present on each plate. When the development of the mites on an individual plate was completed to the desired stage, the plate was removed from the germinator and discarded. All plates were continued through to the emergence of the larvae of the succeeding generation when development to this stage was successful.

The temperature cabinets were maintained at 28 $\pm$ 1°C. As the plates were watered daily, it was necessary to provide some provision for the maintenance of relatively stable humidity conditions. Thus, two large dishes of calcium chloride were placed at the bottom of the interior of the cabinet to absorb excess moisture (Fig. 4). The calcium chloride effectively maintained the relative humidity between 50 and 70 per cent.

While in the cabinet, the light source for the detached leaves consisted of a single 100 watt lamp which automatically illuminated the cabinet between 7 A.M. and 7 P.M. daily. The light was located outside and slightly above the window in the cabinet door (Fig. 4). Light meter readings made with a Weston Illumination Meter,

FIGURE 4

Constant temperature cabinet with experimental plates.

Model 756 (quartz filter), indicated the illumination on the leaf surface approximated 28 foot-candles in the front of the cabinet grading to 5 foot-candles in the rear corners of the cabinet.

The plates of a complete experiment or replicate were placed on a single tray in the cabinet. Previous work had indicated that the position of an experimental plate on the tray within the cabinet did not significantly affect the fecundity of the mites. Nevertheless, it was considered a good general practice to arrange two plates in such a way as to minimize any possible effect of position upon the results of the experiment. With this in mind the plates were arranged in rows of five or six, row 1 being located in the front of the cabinet and row 4 at the back.

In the Nitrogen series each nutrient treatment was replicated in each row. In order to minimize any differences which might exist along the row, plates representing a particular nutrient level were placed at different positions in each row. Method A plates were placed in one cabinet, while Method B plates were placed in another cabinet. This experiment was initiated on October 3, 1959.

The Nitrogen x DDT (detached) series consisted of a total of twenty-four treatments. Thus, a single replicate covered an entire tray and filled the cabinet. Therefore,

the replication in this experiment was done in a time series. The first replicate was started on November 11, 1960, and the other three replicates followed at biweekly intervals. Two cabinets were utilized for the experiment. In this experiment each row contained plates from six different DDT treatments, placed at random within the row. Each nutrient level was represented at least once and not more than twice along the row. Furthermore, two replicates were rotated 180°, thus reversing the position of the rows from back to front and from right to left. Thus, any variable environmental effects within the cabinet were minimized and/or equalized by the distribution of the plates.

Intact leaf procedure.

Nitrogen x DDT (intact) series. In order to ascertain the effects of nitrogen, a single experiment including four replicates was deemed sufficient as there were four replicates available for each treatment level. However, to determine the effects of DDT in the sand at the various nitrogen levels it would be necessary to replicate the experiment in a time series by running four tests on the same plants over an extended period of time. Unfortunately, although the former goal was completed, circumstances restricted the investigation of the effects of DDT to a single replicate.

The first fully formed leaf below the apical meristem was the test leaf. This leaf was isolated from the rest of the plant by means of a one-half inch barrier of petrolatum applied around the petiole to restrain the mites. To prevent the mites from dropping directly to a lower portion of the plant, a card, smeared with petrolatum and supported by a twisted pipe cleaner, was placed directly below the test leaf. The test leaf was not allowed to come into contact with any other portion of the plant, or with the adjacent plant in the crock.

Similar leaves on both plants in each crock were isolated for mite introduction. Thus, one leaf was used for the final counts at the end of the test period, while the other leaf was used to make counts of developmental stages and numbers of mites after a two week period. Under the prevailing temperature conditions, it was estimated that two weeks would be sufficient time to allow any developmental differences which might occur to be expressed.

Mites were introduced on April 15, 1960, and remained undisturbed until April 29 when the first set of leaves was removed and the numbers of mites and their developmental stages recorded. On May 12 and 13, the second set of leaves of the nutrient treatment foliage was removed, the mites counted, and their stages recorded. Although this work spanned two calendar days, the foliage removal

and counting was done within a single twenty-four hour period. On May 12, 13, and 14 (within a 48 hour period) the leaves from the DDT treatment group were removed and similar data recorded.

The crocks were placed in a randomized block design. Each block consisted of ten crocks placed at regular intervals along a twenty-foot plank supported by saw-horses (Fig. 1a). Thus, it was possible to analyze the results of the experiment on the basis of any differential environmental factors due to location of the crocks within the experimental plot.

The temperature range within the experimental plot during the experimental period was 52 to 102°F., with a mean temperature for the period of 73°F. While this temperature is not conducive to rapid population increase, it is suitable for normal T. telarius activity (Whitcomb, 1933; Cagle, 1949). The relative humidity ranged from 20 to 100 per cent with a mean of 65 per cent. The greenhouse was normally maintained at a night temperature of 65°F.

Liquid Nitrogen series. As this series was designed to test the biotic potential of mites under conditions similar to those of commercial greenhouses, no attempt was made to prevent access of stray mites to the test plots. Relying upon natural infestation in short-term experiments,

however, often results in some plots which are not infested during the experimental period. In order to insure that all plots would have countable populations, two young adult female mites were placed on each plant on July 11. At this time two of the four replicates had very light natural mite infestations, while no mites were detected on the other two replicates.

During the experimental period the mite populations were estimated by counting the number of adult female mites on two, three, or five similar leaves per plant. This sampling was done on the four outer-most plants of each plot; the four interior plants remained undisturbed.

Two of the interior plants of each plot were selected for final sampling in September. A total of fifteen leaves was taken from each selected plant: five each from the top, middle, and base of the plant. This sample represented approximately 30-50 per cent of the total number of leaves on a plant. Each leaf was then cut into sections of appropriate size for easy passage between the brushes of the Henderson-McBurnie mite brushing machine (Henderson and McBurnie, 1943).

The mite brushing machine was used to brush the mites from the foliage onto a glass disc coated with sticky varnish. The discs were then refrigerated and stored until the mites could be counted. A radial counting grid

was utilized, and the mites on one-eighth of the surface of the disc were counted with the aid of a stereoscopic microscope. On discs with large numbers of mites one-sixteenth of the surface area was surveyed, and on discs with fewer mites one-quarter of the disc was surveyed. An appropriate multiplication factor was then used to give the approximate numbers of mites for the entire disc, and thus for the fifteen leaf samples. Arthropod predators were also counted.

FOLIAR ANALYSIS

General.

Foliage samples were generally taken from the plants late in the day. In order to conserve time samples were taken throughout the day in the Liquid Nitrogen series. The samples were placed in paper bags and dried in a ventilated oven at 70°C. for three days, after which they remained in the bags until being ground. Prior to grinding the foliage in a Wiley mill (intermediate model), they were again dried at 70°C. for a twenty-four hour period. A twenty mesh screen was used in the Wiley mill. The samples were then stored in bottles or vials, and were dried again prior to weighing for chemical analysis.

Nitrogen.

All nitrogen determinations were done in duplicate by the author using the micro-Kjeldahl technique of Stubblefield and DeTurk (1940) reduced roughly ten times in the amounts of chemicals used. Depending upon the availability of foliar material, determinations were made with 50, 100, or 200 mg. samples. Kjeldahl nitrogen is the total nitrogen in the foliage expressed as the percentage of the dry weight of the foliage.

Vermiculite series. Three groups of plants were analyzed for nitrogen: those harvested after sixteen weeks, seventeen weeks, and eighteen weeks.

The sixteen week group (I) was analyzed in the following manner. The foliage of each plant was divided into upper leaves (the top four leaves of each stem) and lower leaves (the remaining leaves on the plant with the exception of the two basal leaves of each stem). The foliar samples of each plant were analyzed individually. There was so little variation between the replicates within the treatments that they were combined in the remaining groups. Thus, composite treatment samples were utilized in both the seventeen (II) and eighteen (III) week groups.

The apical meristems were prepared in the same manner as was the foliage. The meristems from all groups were combined into composite samples for each treatment level

for chemical analysis.

Nitrogen series. Foliage samples were taken approximately three weeks before leaves were detached for experimental purposes. A single leaf, similar in age and position to the test leaves, was taken from each of the four replicate crocks at each nitrogen treatment level; these leaves were combined into one composite sample for that level. Separate samples were made for both Method A and Method B.

Nitrogen x DDT (detached) series. Samples were taken from the non-DDT treatments. The top four fully-formed leaves from the upper shoot in each crock were combined into composite samples for each nitrogen level. Although the experimental work included only the upper four nitrogen treatment levels, all five levels were analyzed for nitrogen content of the foliage.

Nitrogen x DDT (intact) series. During the test period several new leaves were produced above the test leaf due to plant growth. The foliage on the stem between the starting position and the final lowest position of the test leaf was considered to be representative of the nutritional status of the test leaf during the test period. Thus, at the end of the experimental period the foliage between the test leaf and the apex of the plant was analyzed for nitrogen content. Each replicate was analyzed individually.

Liquid Nitrogen series. The entire foliage from each plant sampled for mite population totals was removed for analysis. In some cases the foliage from each individual plant was bagged separately, while in other cases the foliage from the two plants in one plot was combined. In the former case the individual samples were analyzed and the results averaged, while in the latter instance the combined sample was analyzed.

Ca, Mg, P and K.

Chemical analyses of the foliage for calcium, magnesium, phosphorus, and potassium were made by the Regulatory Service, Massachusetts Agricultural Experiment Station, Amherst, Mass. Phosphorus was determined colorimetrically with a Bausch & Lomb Spectronic 20 spectrophotometer (Gersten, 1957). Potassium was determined by flame photometry employing a Beckman DU Spectrophotometer using the A.O.A.C. method (1960), with slight modification. Calcium and magnesium were determined by titration with Versenate (Cheng et al., 1952).

For these analyses composite samples were submitted. From the Vermiculite series, the foliar and meristematic tissue was combined to give a single sample for each nutrient treatment level. The foliage from single stems was combined into composite samples for both Method A and

Method B of the Nitrogen series. Similarly, the foliage from each treatment in the Liquid Nitrogen series was combined to make composite samples for each of the nine treatments and the check (no nitrogen-no DDT).

SOIL ANALYSIS

Soil analyses were made by station personnel under the direction of Dr. Norman E. Butterfield, Waltham Field Station, University of Massachusetts, Waltham, Mass. Standard Morgan soil testing techniques were used (Lunt et al., 1950).

The soil was sampled by mixing several trowels-full of soil from the central position of each plot (the area of the plants which received the final sampling). After thorough mixing, one cup of soil was submitted for analysis.

STATISTICAL ANALYSIS OF DATA

All mite counts reported herein, unless otherwise specified, include both eggs and active stages. The data for developmental periods were determined by observation of the first individual to reach a specified stage and thus is a measure of the minimum period of development starting from oviposition.

Standard methods were used for analysis of variance (single and multiple classifications), correlation, and regression (Snedecor, 1956; Cochran and Cox, 1957).

RESULTS OF INVESTIGATION

The data which form the bases for the graphs presented in this section have been tabulated and, in most cases, statistically analyzed. The individual data, means, analyses of variance, least significant differences, and b values are located in Tables 5 through 19 in the Appendix. Specific reference to these tables will be made in the body of the text to co-ordinate graphic presentation with original data.

PLANT RESPONSE TO NITROGEN

Vermiculite series.

Plant response, in terms of foliar nitrogen content, is shown in Figure 5 and Table 5. A highly significant linear response was found in both the upper and lower leaves of Group I. Similar responses were apparent in Groups II and III, indicating that while absolute values varied slightly from week to week, the general response was consistent. The meristematic regions had higher nitrogen contents at the lower levels of nitrogen application than did either the upper or lower leaves, but little difference was found at the higher levels of application.

The mean measurements from twelve plants at each

nitrogen level are presented in Table 4. The entire plant was utilized for the weight data, while the remaining measurements were taken from the upper stem.

Table 4. Mean plant growth response to nitrogen treatments.

Nitrogen level (ppm.)	Fresh Weight		Height	Number of	Internode length
	foliage (grams)	stems (grams)	(Inches)	leaves	(inches)
25	5.9	5.8	10.1	9.1	1.11
50	10.7	9.5	13.0	11.2	1.16
100	16.4	12.3	14.4	13.2	1.09
200	18.8	13.5	15.1	14.4	1.05
400	16.4	13.2	13.2	14.3	0.92

Foliage at the lower nutrient levels (25, 50 ppm.) was small, tough, and pale; at the intermediate level (100 ppm.) leaf size, texture, and color were normal; and at surplus levels (200, 400 ppm.) the foliage was dark, large, and succulent. Maximum root growth occurred at the 100 ppm. level, while higher or lower levels of nutrient application produced smaller roots. These observations coincide with the data in Tables 4 and 5 in indicating that the 100 ppm. level produced normal growth, higher levels of nitrogen application stimulated plant growth, and lower levels resulted in stunted plants.

Sand culture series. (Nitrogen and Nitrogen x DDT (detached and intact) series.)

Plant response was similar to that reported above for the Vermiculite series. Chemical analyses showed that the nitrogen content of foliage produced under Method A was substantially less than that of Method B at the 100, 200, and 400 ppm. levels (Figure 6). Comparison of Figures 5 and 6 indicates that the foliar nitrogen content with Method A at these levels was essentially the same in vermiculite and sand culture.

Liquid Nitrogen series.

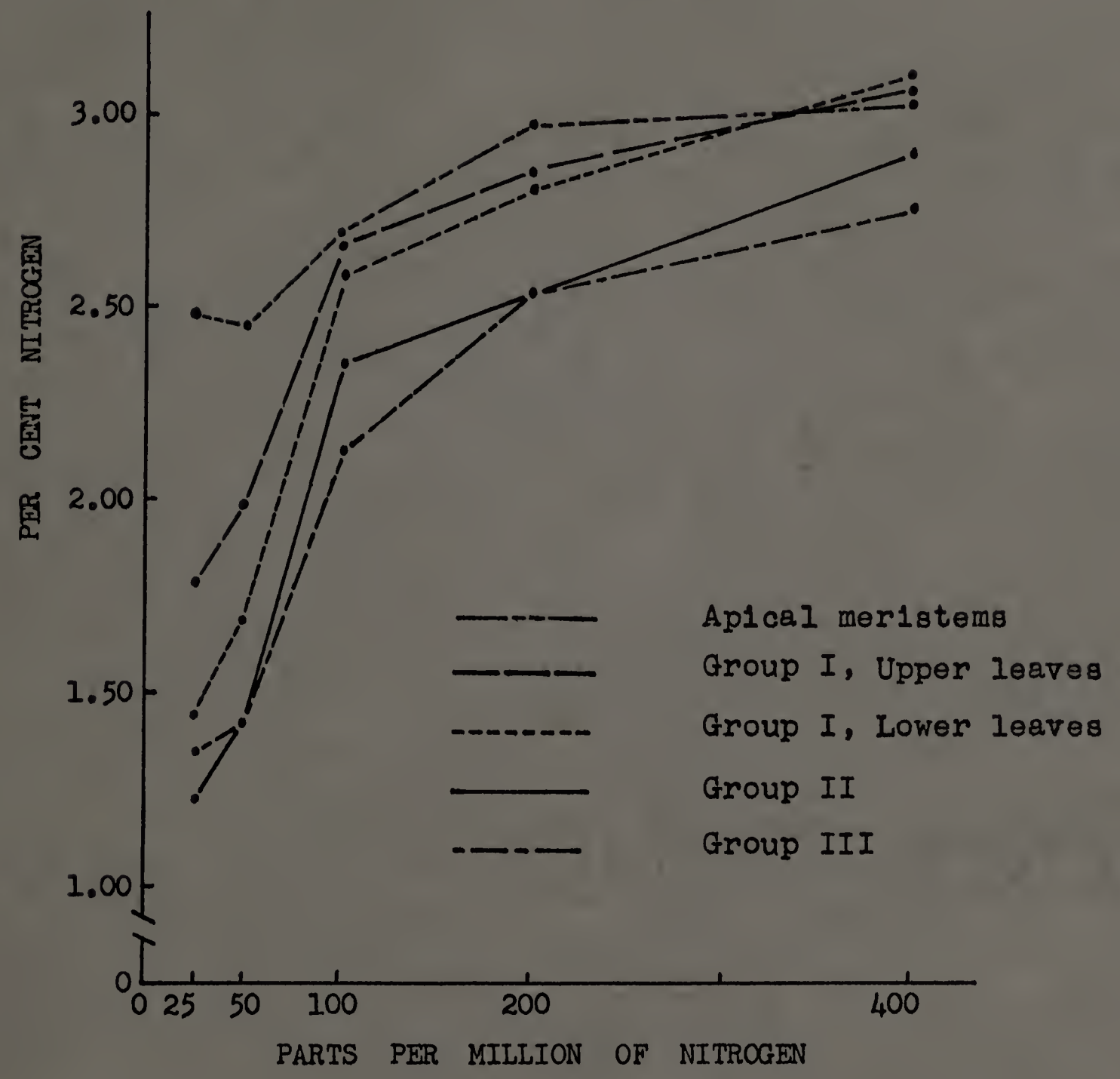
There were no significant differences in plant height or number of leaves among the treatments receiving liquid nitrogen. However, the average height of the check plants (0 ppm.) was 15 per cent less than that of the plants which received supplementary nitrogen. The nitrogen content of the foliage showed a positive and highly significant linear response to increased nitrogen application (Table 16).

Ca, Mg, P, and K.

While statistical analyses cannot be applied to the data from the chemical analysis of the composite samples (Table 19), inspection of these data indicates that potassium was absorbed concomitantly with nitrogen. Absorption

FIGURE 5

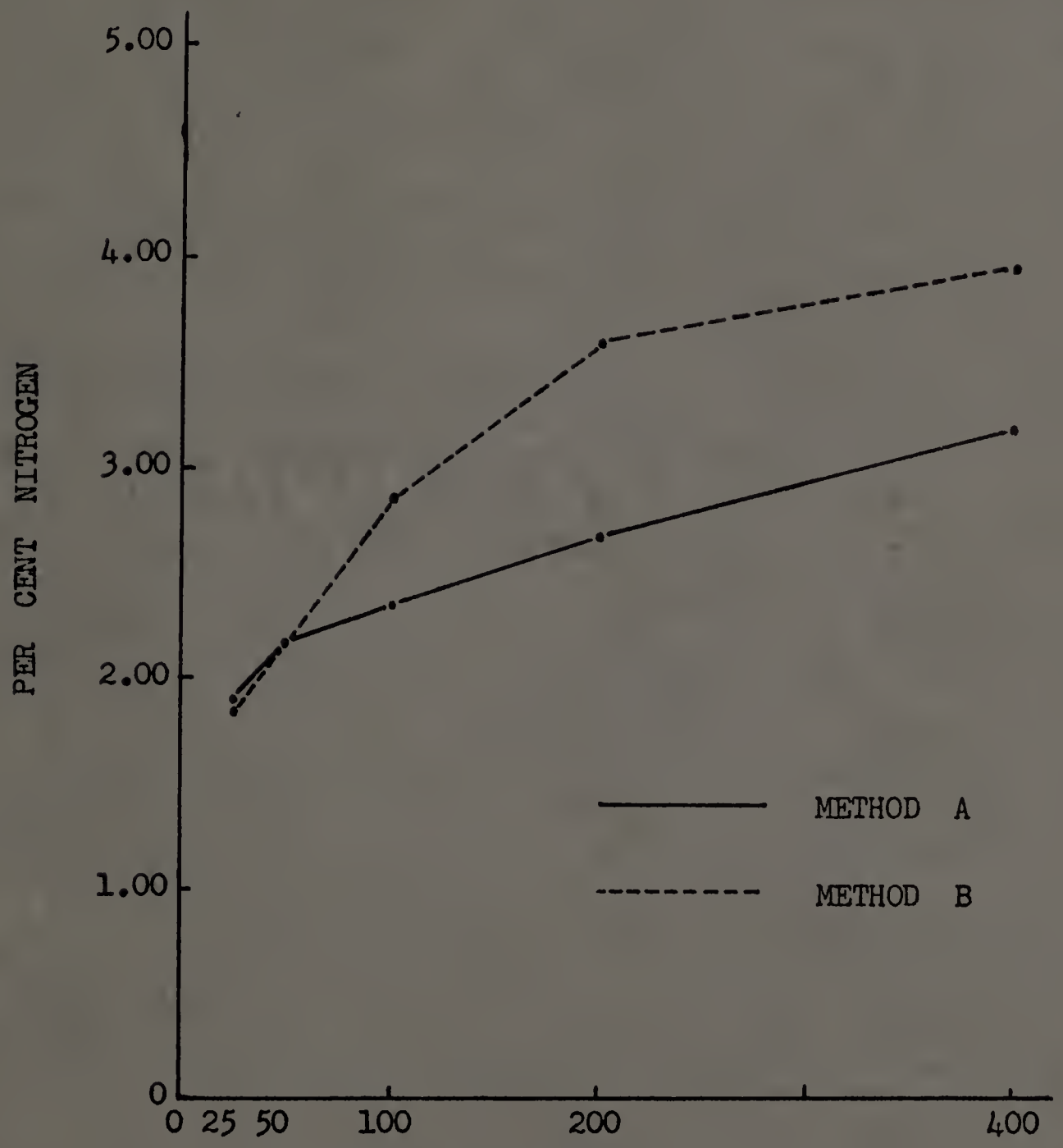
NITROGEN CONTENT OF FOLIAGE AT FIVE NITROGEN LEVELS *



* Foliage from chrysanthemums grown in Terralite. Nitrogen increases in nutrient solutions made by varying the amounts of $Ca(NO_3)_2$, KNO_3, KCl, $CaCl_2$. Nitrogen content expressed as per cent of dry weight of foliage. Samples taken at: 16 weeks (Group I); 17 weeks (Group II); 18 weeks (Group III).

FIGURE 6

NITROGEN CONTENT OF FOLIAGE AT FIVE NITROGEN LEVELS *



* Foliage from chrysanthemums grown in sand culture. Nitrogen content expressed as per cent of dry weight of foliage. Nitrogen increases in nutrient solutions made by:

METHOD A: varying amounts of $Ca(NO_3)_2$, KNO_3, KCl, $CaCl_2$

METHOD B: addition of NH_4NO_3.

of magnesium was quite variable; calcium uptake did not seem to be affected. Phosphorus, which was variable in the Vermiculite and Nitrogen series, appeared to be absorbed concomitantly with nitrogen in the Liquid Nitrogen series.

PLANT RESPONSE TO DDT

Plant response to DDT was measured in terms of foliar nitrogen content. Determinations were made for the Nitrogen x DDT (intact) and Liquid Nitrogen series.

A strict comparison between the DDT and non-DDT treatments of the Nitrogen x DDT (intact) series is not feasible because the former group was replicated only once. In general, however, there was little difference in the nitrogen content of the DDT and non-DDT groups (Tables 12 and 15B). DDT had no effect on the foliar nitrogen content in the Liquid Nitrogen series either as a spray or a soil treatment (Table 16).

MITE RESPONSE TO NITROGEN

Nitrogen series.

Leaves detached from chrysanthemums grown in sand culture at various nitrogen levels were utilized in this

experimental series. Mite fecundity showed a highly significant linear response to increases in nitrogen supplied to the plant (Figure 7, Table 6). Nutrient Methods A and B gave similar linear responses, showing that mite fecundity was enhanced by increasing amounts of nitrogen. However, only Method B yielded significant differences among treatments.

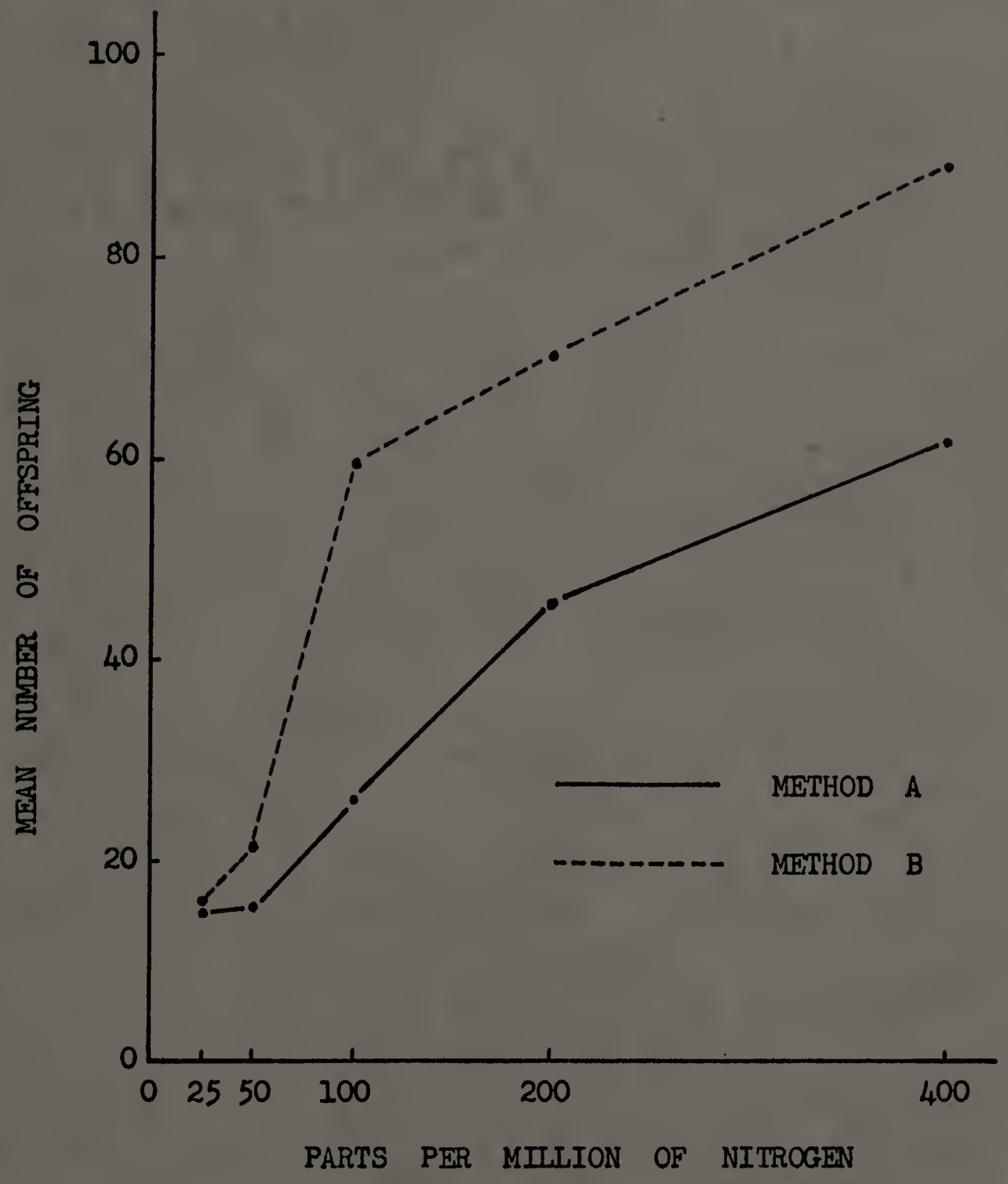
Figure 8 and Table 7 show the mite response in terms of developmental rate. The developmental period was significantly reduced at the higher levels of nitrogen application with Method A; Method B gave results which approached significance. Both methods produced highly significant linear responses to the increasing nitrogen applications.

The lack of significant differences between treatments in this experiment was due to the high degree of individual variation displayed by the mites. Thus, even though the trends were obvious and the extremes widely separated, significant differences were not found where casual perusal of the data might lead one to expect them.

The difference in mite response which exists between nutrient Methods A and B closely parallels the difference in the nitrogen content of the foliage produced by the two methods (Figure 6). Method B was more suitable for mite fecundity and development. It is not known what part, if any, either chlorine or high nitrate concentration played

FIGURE 7

MEAN NUMBERS OF OFFSPRING OF THREE ADULT FEMALE MITES AT FIVE NITROGEN LEVELS *



* Reared on leaves excised from chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by:

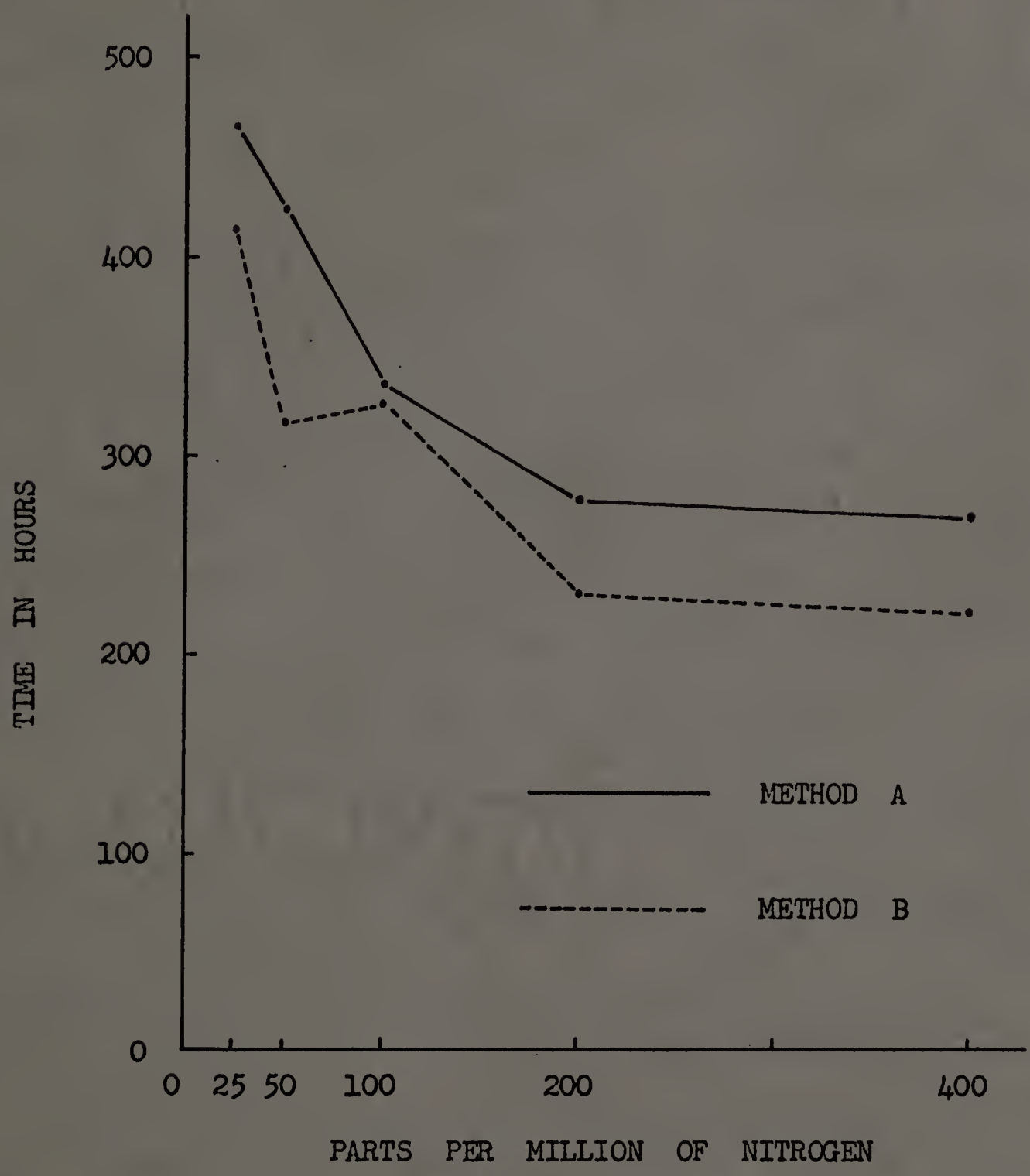
METHOD A: varying amounts of $Ca(NO_3)_2$, KNO_3, KCl, $CaCl_2$

METHOD B: addition of NH_4NO_3.

FIGURE 8

MEAN DEVELOPMENTAL PERIODS (EGG TO ADULT FEMALE MITE)

AT FIVE NITROGEN LEVELS *



* Reared on leaves excised from chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by:

METHOD A: varying amounts of $Ca(NO_3)_2$, KNO_3, KCl, $CaCl_2$

METHOD B: addition of NH_4NO_3.

in producing the relatively lower foliar nitrogen concentration resulting from Method A. It is noted, however, that at the 400 ppm. level (Method A) and the 100 ppm. level (Method B) both the foliar nitrogen content and the mean number of offspring are very similar. Thus, with either method the reproductive response of the mites at a given level of foliar nitrogen appears to be the same.

MITE RESPONSE TO NITROGEN AND DDT

Nitrogen x DDT (detached) series.

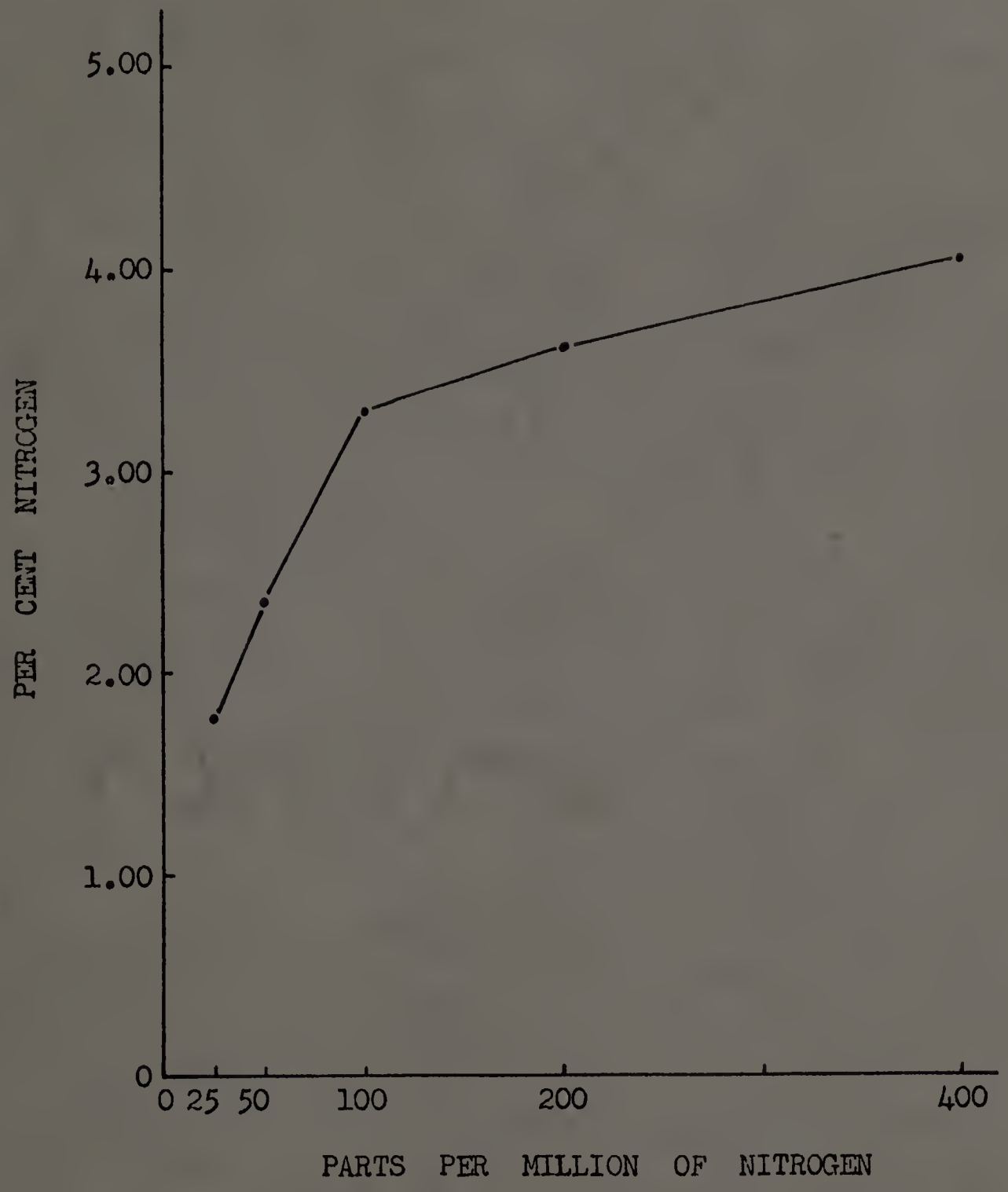
Leaves detached from plants grown in sand culture at various nitrogen and DDT levels were utilized in this series. Nutrient Method B was used.

Figure 9 shows the foliar nitrogen content of the non-DDT treatments. The mean numbers of offspring resulting from the various nitrogen and DDT treatments are presented in Figure 10 and Table 8.

The effect of nitrogen on fecundity is illustrated in Figure 11. Although the 50 and 100 ppm. levels of nitrogen application were not significantly different, highly significant differences were found between the 100 and 200 and the 200 and 400 ppm. levels. The positive response to increased nitrogen showed a highly significant degree of linearity.

FIGURE 9

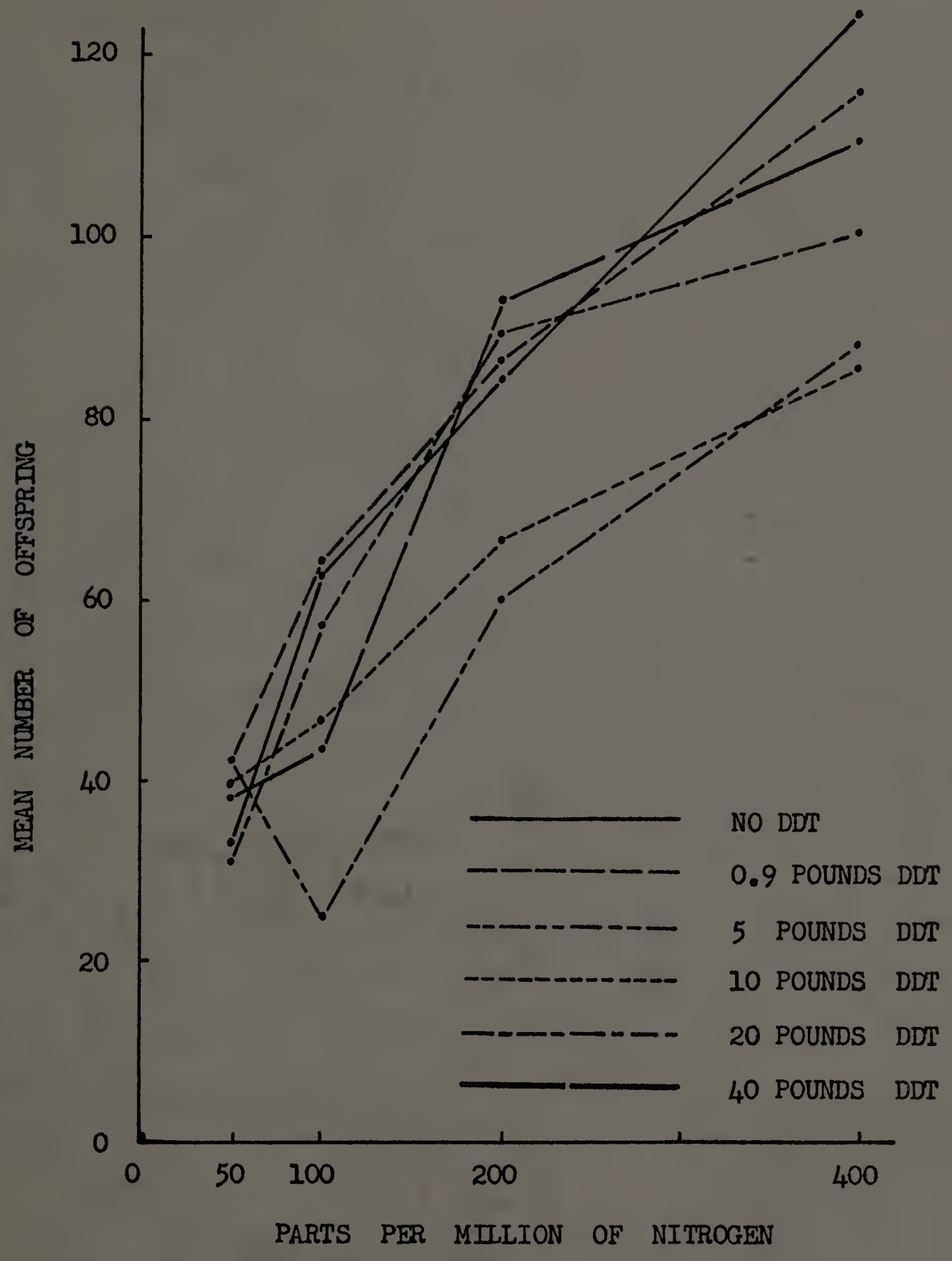
NITROGEN CONTENT OF FOLIAGE AT FIVE NITROGEN LEVELS *



* Foliage from chrysanthemums grown in sand culture. Nitrogen content expressed as per cent of dry weight of foliage. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3.

FIGURE 10

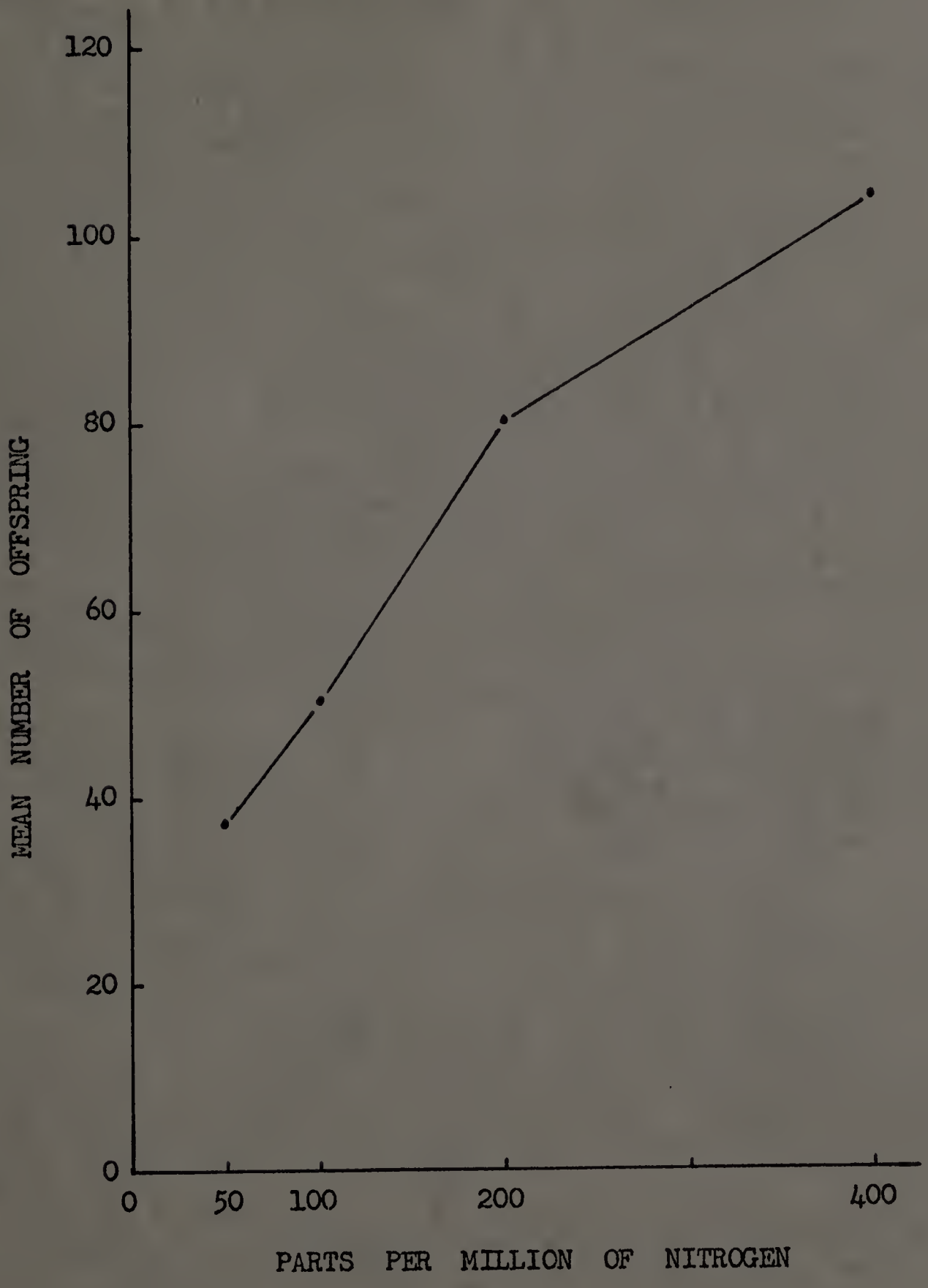
MEAN NUMBERS OF OFFSPRING OF THREE ADULT FEMALE MITES AT FOUR NITROGEN LEVELS AND SIX DDT LEVELS *



* Reared on leaves excised from chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3. DDT applied to sand @ 0, 0.9, 5, 10, 20, 40 pounds per acre.

FIGURE 11

EFFECTS OF NITROGEN ON MEAN NUMBERS OF OFFSPRING OF THREE ADULT FEMALE MITES AT SIX DDT LEVELS *



* Reared on leaves excised from chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3. DDT applied to sand @ 0, 0.9, 5, 10, 20, 40 pounds per acre.

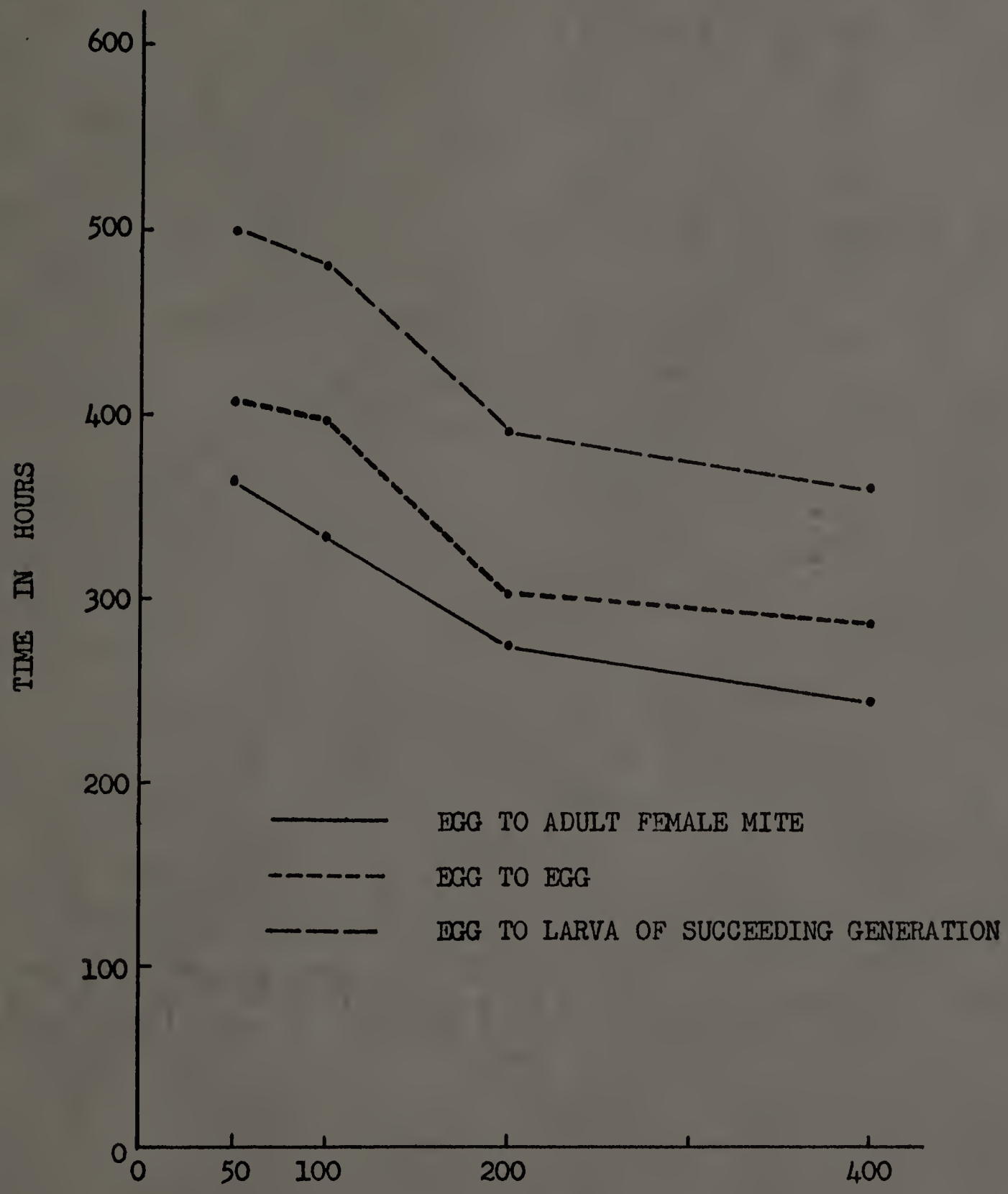
The developmental rate (Figure 12 and Tables 9, 10, and 11) increased with increasing nitrogen supply. Highly significant differences were found between the 100 and 200 ppm. levels. Again the linear response was highly significant, although a significant degree of deviation from linearity was noted in the egg to egg and egg to larva of succeeding generation data. These deviations from linearity, not uncommon in biological systems, indicate that the rate of stimulation decreases with increasing amounts of stimulant.

The 5 pounds of DDT per acre data were excluded from the analysis of variance of the developmental periods to the egg and larval stages because of the failure of the mites to reach these stages at the 50 ppm. level of nitrogen application.

Although DDT had no apparent effect upon the rate of development, the 5 and 10 pound rates significantly reduced fecundity (Figure 13). Close inspection of the data shows that this reduction occurred only at normal and surplus levels of nitrogen application, and not at levels where fecundity was already depressed by insufficient nutrients. The drop in fecundity assumed a curve not unlike that of a quadratic response and gradual recovery to normal was apparent at the 20 and 40 pound levels.

FIGURE 12

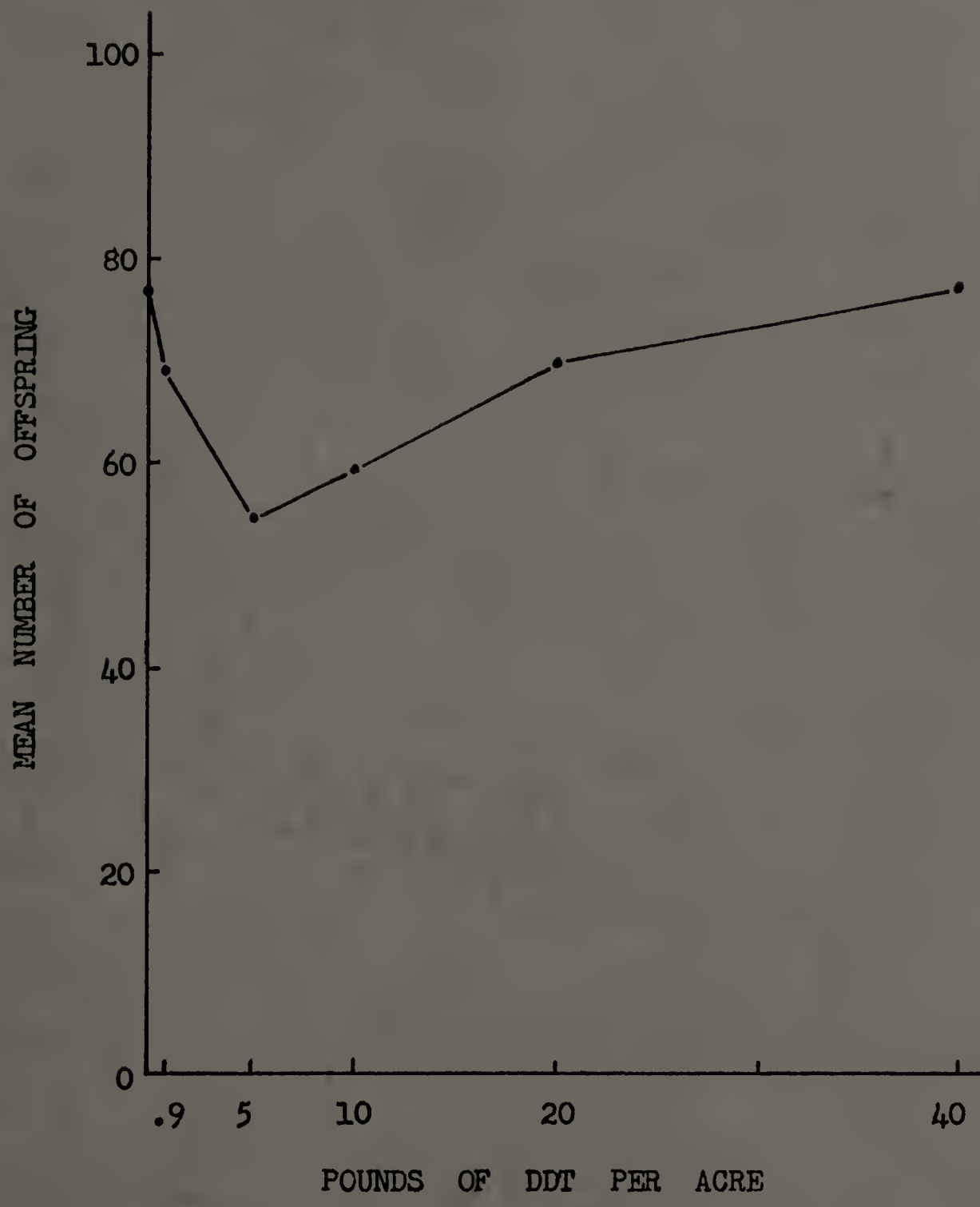
EFFECTS OF NITROGEN ON MEAN DEVELOPMENTAL PERIODS AT SIX DDT LEVELS *



* Reared on leaves excised from chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3. DDT applied to sand @ 0, 0.9, 5, 10, 20, 40 pounds per acre; five pound per acre data incorporated in egg to adult curve only.

FIGURE 13

EFFECTS OF DDT ON MEAN NUMBERS OF OFFSPRING OF THREE ADULT FEMALE MITES AT FOUR NITROGEN LEVELS *



* Reared on leaves excised from chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3. Nitrogen levels: 50, 100, 200, 400 parts per million.

Nitrogen x DDT (intact) series.

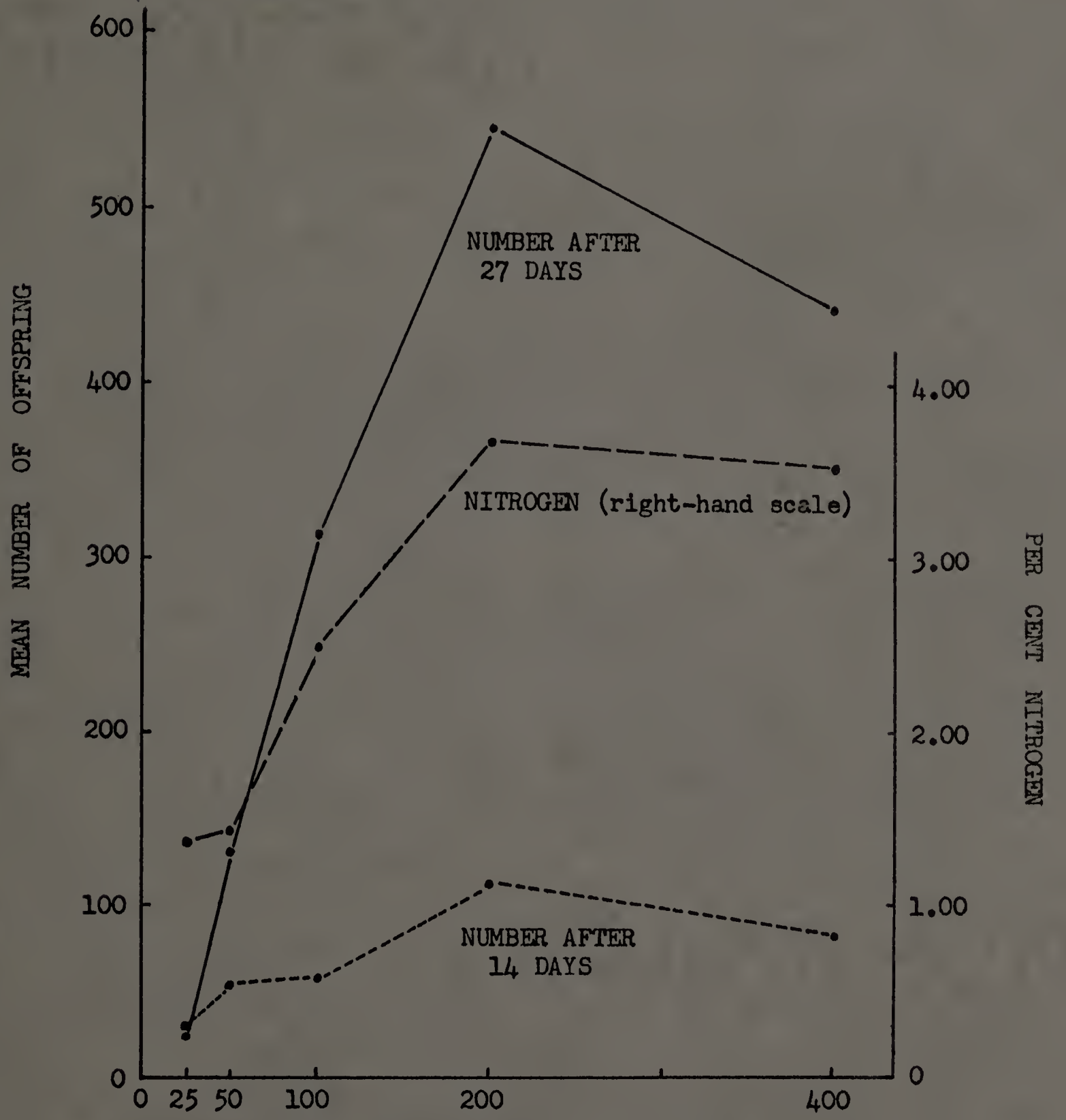
Isolated, intact foliage on chrysanthemums grown in sand culture at various nitrogen and DDT levels was utilized in this series. Nutrient Method B was used; the volume applied weekly was increased by 70 per cent over that of the Nitrogen x DDT (detached) series.

Figure 14 and Tables 13 and 12 show the mean numbers of offspring after 14 and 27 days and the mean nitrogen content of the foliage of the non-DDT treatments. The numbers of offspring for both periods and the foliar nitrogen content revealed highly significant differences among the nitrogen treatments. Increases in offspring and foliar nitrogen content showed highly significant linear responses to increases in nitrogen application. The drop in fecundity at the 400 ppm. level coincided with a similar drop in foliar nitrogen. Figure 15 shows a highly significant positive correlation between foliar nitrogen and the number of offspring after 27 days. The regression coefficient reveals an average increase of 175 mites per one per cent rise in foliar nitrogen with a standard deviation of 31.

An antibiotic effect was apparent at the 25 ppm. level at which there was no increase in the number of offspring between the 14 and 27 day counts.

FIGURE 14

NITROGEN CONTENT OF FOLIAGE AND MEAN NUMBERS OF OFFSPRING OF THREE ADULT FEMALE MITES AT FIVE NITROGEN LEVELS *



* Reared on chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3. Nitrogen content expressed as per cent of dry weight of foliage.

FIGURE 15

* Reared on chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3. Nitrogen content expressed as per cent of dry weight of foliage.

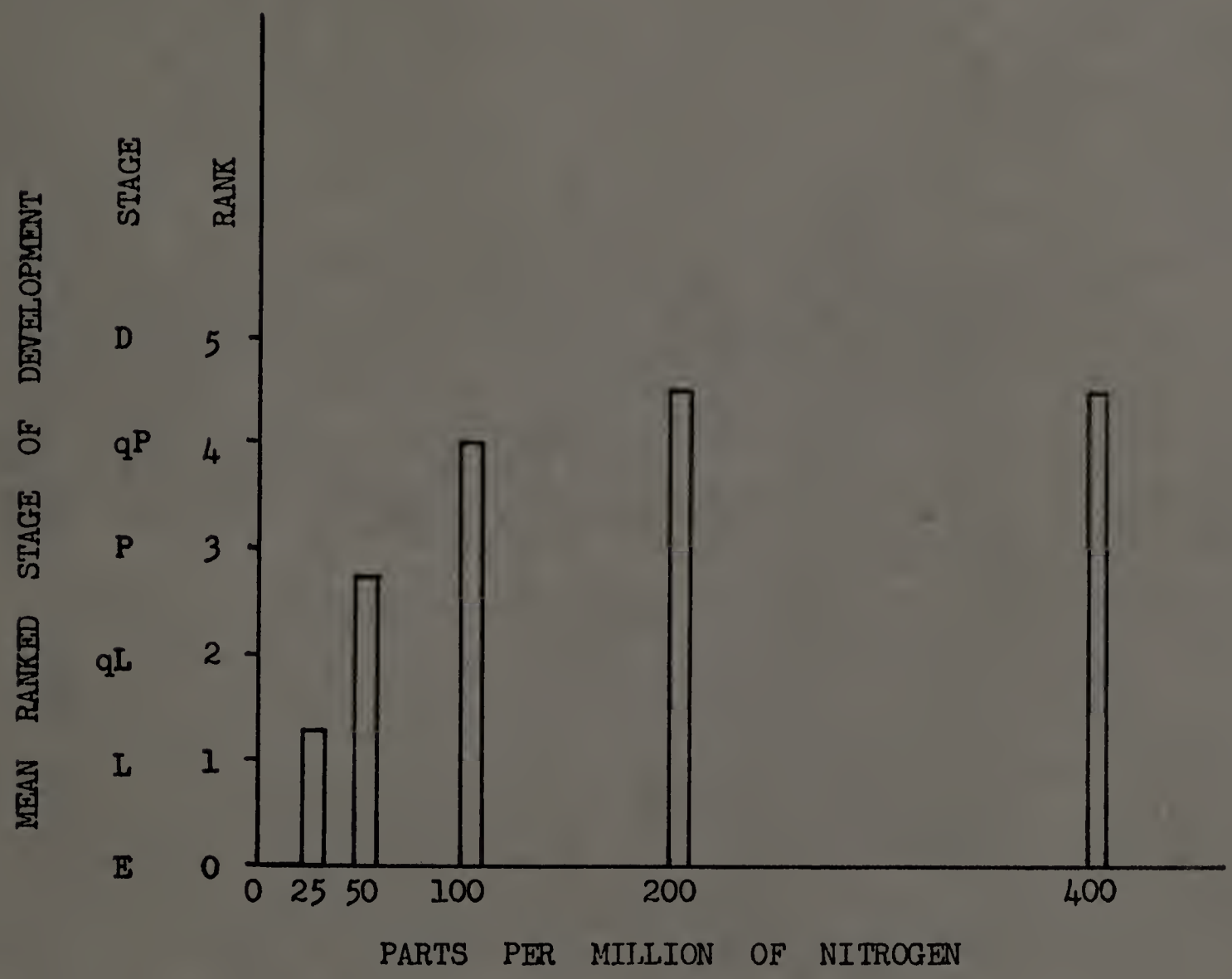
The rate of development (Figure 16, Table 14) was also found to increase with increasing nitrogen application (and foliar content) and exhibited a highly significant linear response to treatment. Slower developmental rates in this series than in the Nitrogen x DDT (detached) series were due primarily to lower prevailing temperatures.

The results of the DDT treatments are given in Table 15. This portion of the experimental series was not completed and data from a single replicate can be analyzed only with caution.

Of particular interest is the apparent increased fecundity of the mites on DDT-treated plants, in comparison to the non-DDT plants, at the 100, 200, and 400 ppm. levels of nitrogen application. In addition, the developmental rate appeared to be more rapid at the 200 and 400 ppm. levels on the DDT-treated plants than on the non-DDT group. While it is indeed unfortunate that these treatments were not replicated, the available data indicate that the DDT may have enhanced the biotic potential of the mite. This apparent stimulation does not seem to be connected with the nitrogen content of the foliage.

FIGURE 16

MEAN STAGE OF DEVELOPMENT OF MITES AFTER FOURTEEN DAYS AT FIVE NITROGEN LEVELS *



* Reared on chrysanthemums grown in sand culture. Nitrogen increases in nutrient solutions made by addition of NH_4NO_3.

D = deutonymph; qP = quiescent protonymph; P = protonymph; qL = quiescent larva; L = larva; E = egg.

Liquid Nitrogen series.

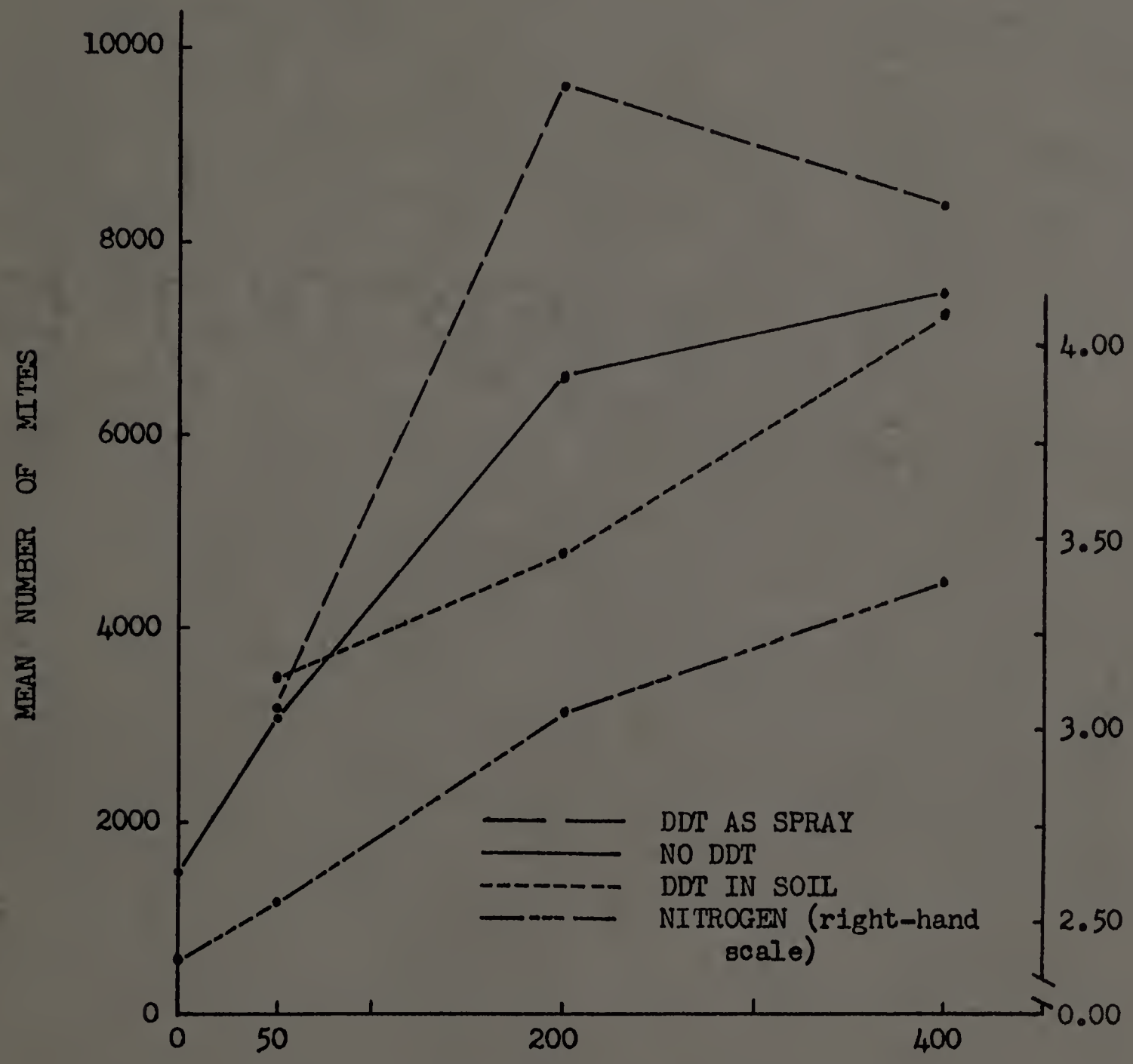
Plants grown under conditions which approached those of commercial operations were utilized for this series.

Figure 17 and Tables 16 and 17 indicate the mite populations and foliar nitrogen levels which developed with the various nitrogen and DDT treatments. There was a highly significant linear response to increasing levels of nitrogen application in terms of both foliar nitrogen and mite populations. The surplus nitrogen treatments (200, 400 ppm.) produced mite populations which, while not significantly different from each other, were significantly higher than the normal (50 ppm.) nitrogen treatment. Figure 18 shows a highly significant positive correlation between foliar nitrogen and mite populations. The regression coefficient indicates an average increase of 362 mites per one per cent increase in foliar nitrogen with a standard deviation of 60.

Neither soil nor foliar applications of DDT significantly affected mite populations or foliar nitrogen content. However, the analysis of mite populations showed a significant interaction between DDT and nitrogen. This interaction occurred at the surplus levels of nitrogen application and thus does not affect the results stated above

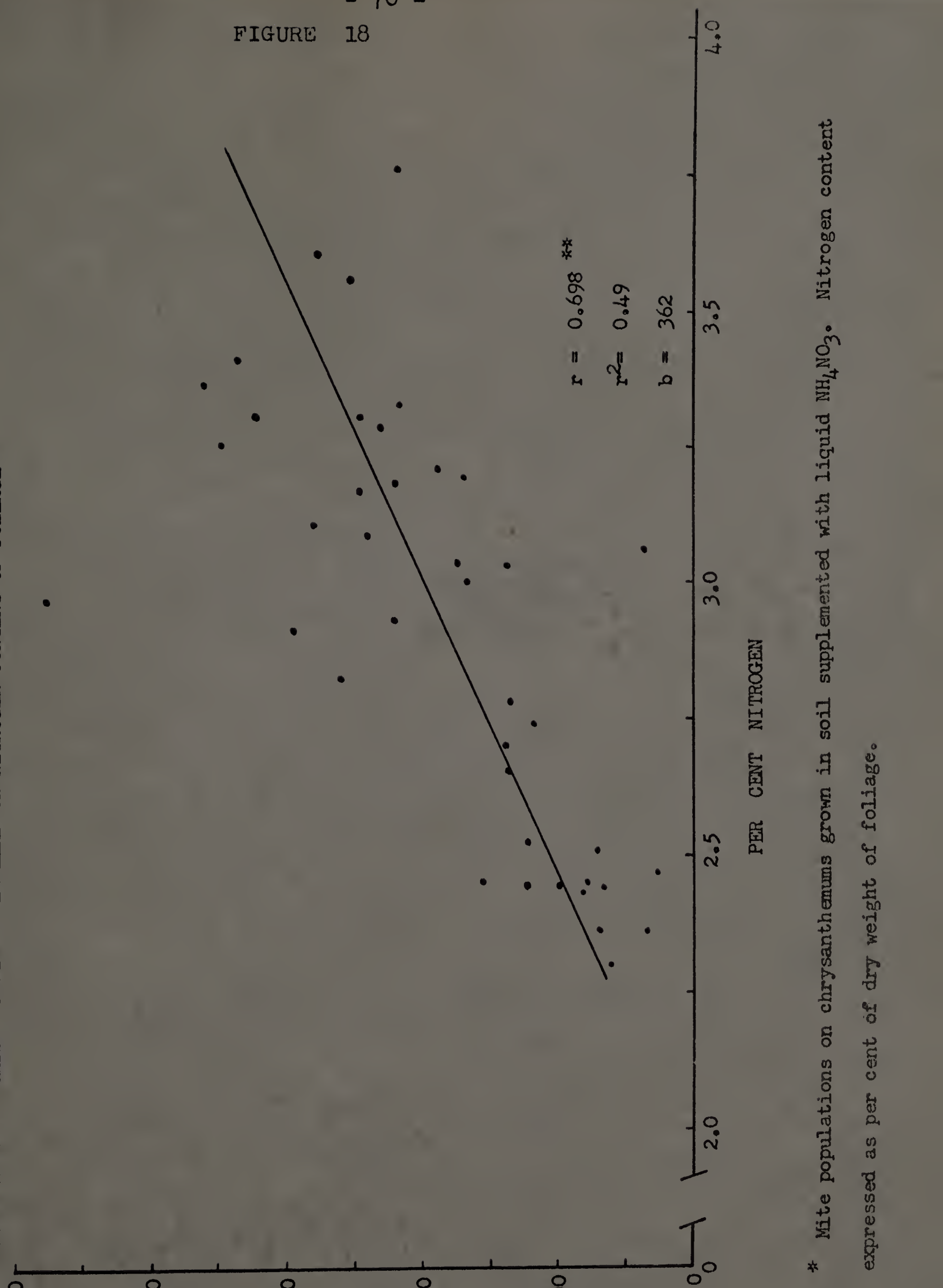
FIGURE 17

MEAN NUMBERS OF MITES PER FIFTEEN LEAVES AT FOUR NITROGEN LEVELS AND MEAN NITROGEN CONTENT OF FOLIAGE WITH THREE DDT TREATMENTS *



* Mite populations on chrysanthemums grown in soil supplemented with liquid NH_4NO_3 applications. No DDT treatment at zero nitrogen level. Nitrogen content expressed as per cent of dry weight of foliage.

FIGURE 18



* Mite populations on chrysanthemums grown in soil supplemented with liquid NH_4NO_3. Nitrogen content expressed as per cent of dry weight of foliage.

because no significant differences were found at these levels.

Unidentified coccinellid, chrysopid, and syrphid predators were recovered by the mite brushing technique. Although not numerous, more insect predators were found on the surplus nitrogen levels than elsewhere. Typhlodromus fallacis (Garman), an important mite predator of T. telarius (Ballard, 1954), was also active during this experiment. Ristich (1956) reported this mite to be highly susceptible to DDT; in this experiment it was less abundant on the DDT sprayed plots than elsewhere at any given nitrogen level. In spite of this distribution, higher T. telarius populations were not consistently correlated with DDT sprayed plots. Furthermore, the 200 ppm. nitrogen treatment had the highest T. fallacis population and the second highest T. telarius population, indicating that the predator mite was not an influential factor in regulating T. telarius abundance. Thus, the presence of natural enemies did not bias the results of the experiment.

Inspection of the soil analysis data (Table 18) indicates that at the highest level of nitrogen application considerable amounts of nitrate nitrogen were not being utilized. These data may partially explain the non-signif-

icant increase in mite populations between the 200 and 400 ppm. nitrogen levels. In addition, the soil pH dropped with increasing levels of nitrogen application. The soil potassium level decreased with increasing nitrogen application, indicating that potassium was being absorbed concomitantly with nitrogen. This finding agrees with the foliar analysis which revealed higher levels of potassium at the surplus nitrogen levels.

DISCUSSION OF THE RESULTS

Individual variation.

One of the major problems encountered in experimental work with T. telarius is a high degree of individual variation. This factor has been recognized by previous workers (Rodriguez, 1958; Garman, 1959), and is particularly evident in terms of fecundity. The effects of individual variation can be minimized in experimental work by increased replication. This approach was taken in these studies (after the Nitrogen series failed in some instances to produce significant results even though the general trends appeared obvious) and resulted in sound statistical evidence to support the conclusions stated at the end of this report.

Plant response.

Evidence was presented in the section dealing with plant response which indicated that chrysanthemums grown in sand and vermiculite produced normal growth at the 100 ppm. level of nitrogen application; lower or higher levels were deemed deficient and surplus, respectively.

As the current greenhouse practice is to feed chrysanthemums at a rate approximating the 50 ppm. level used in the Liquid Nitrogen series (Andrease et al., 1956), this level was regarded as producing normal plants. Plants receiving no additional nitrogen were deficient, and application of over 50 ppm. was regarded as surplus. The accuracy of these categories was confirmed by plant measurements and foliar nitrogen content.

Leaf age.

When the nitrogen supply was limited, new foliage was found to have higher total nitrogen content than old foliage. Because higher nitrogen content has been found to increase mite fecundity, it would appear that the younger foliage would be more suitable for population increases as was reported for P. citri (Henderson and Holloway, 1942; Fleschner, 1952; Jeppson et al., 1957). Although not experimentally tested in these studies, such a correlation would agree with the work by Kennedy and

Booth (1951), which showed that the bean aphid, Aphis fabae (Scopoli), on spindle (Euonymus europaeus) and sugar beet, preferred to feed and reproduced faster on young leaves than on mature foliage. Working with the pea aphid, Macrosiphum pisi (Harris), Maltais and Auclair (1957) found that the terminal growth of peas had higher nitrogen content than the middle growth. Thus, other factors being equal, it seems probable that the natural tendency of T. telarius to move up the plant to new growth would enhance its biotic potential.

DDT.

Although DDT was not found to affect the nitrogen content of the foliage, physiological changes occurred within the plants which altered mite biotic potential. A significant reduction in fecundity was found with mites reared on leaves detached from plants grown in sand treated with 5 and 10 pounds of DDT per acre. Gradual recovery occurred at the 20 and 40 pound levels. This inhibition occurred at normal and surplus levels of nitrogen application, but not at deficient levels. A similar trend towards reduction and subsequent recovery at increased dosages of DDT was noted by Rodriguez et al. (1957).

Fecundity and developmental rate seemed to be stim-

ulated by DDT when mites were reared on the intact foliage of plants grown in treated sand at normal and surplus levels of nitrogen application. This observation adds to the accumulating evidence that physiological changes occurring in the plant due to the presence of DDT are favorable for mite increase. The incomplete nature of the experimental series, however, precludes any generalization based on these observations.

DDT applied to greenhouse soil at 20 pounds per acre did not affect mite populations or nitrogen content of the foliage significantly. As DDT affects plants to a greater degree on light soils and sands than on heavy organic soils (Goldsworthy, 1948; Foster, 1951; Thurston, 1953), one would expect less plant and mite response in soil than in sand culture. Klostermeyer and Rasmussen (1953) and Rodriguez et al. (1960a, 1960b) found that heavy soil application of DDT resulted in mite increases.

Nitrogen.

Fecundity. These studies supply further experimental evidence that increased nitrogen supply enhances mite fecundity. With most plants investigated thus far, a positive correlation between nitrogen and T. telarius was well established before the studies reported herein were initiated. With few exceptions, increased nitrogen

application or foliar nitrogen concentrations were found to result in increased T. telarius populations (See Review of Literature).

Developmental rate. Not heretofore demonstrated is the finding that increased nitrogen application and foliar nitrogen content were significantly correlated with an increase in the developmental rate of T. telarius. This finding is in general agreement with Breukel and Post (1959), who, while unable to establish statistical significance, reported a tendency of P. ulmi to develop at a more rapid rate when there was more nitrogen in the foliage. Hamstead and Gould (1957), however, stated that mite increases (T. telarius and P. ulmi) correlated with increased foliar nitrogen were due to an increase in fecundity and that the rate of development was not affected.

General.

It is apparent that increased nitrogen application is the triggering mechanism which initiates plant changes resulting in increased foliar nitrogen and enhanced mite biotic potential. Although significant correlations repeatedly have been found between nitrogen and mite populations, it cannot be assumed that it is the foliar nitrogen, per se, which causes the stimulation.

Several workers have found that increased nitrogen applications result in increased foliar carbohydrate concentration (Rodriguez *et al.*, 1960b; Henneberry and Smith, 1960). Fritzsche *et al.* (1957) correlated *T. telarius* increases with carbohydrate, glutamin, and glutamic acid increases in the foliage. It is expected that future studies will probe more deeply into the basic relationship which exists between *T. telarius*, its host plants and environment.

CONCLUSIONS

Previous workers have indicated that the relationship which exists between Tetranychus telarius (Linnaeus) and its host plant can be altered by varying the amount or type of nitrogen supplied to the plant. The investigations reported herein were designed to study the effects of nitrogen and DDT upon the relationship between T. telarius and chrysanthemums. Based upon the experimental conditions described in this report, the following conclusions are drawn.

Chrysanthemums responded to variation in nitrogen supply; increasing amounts of nitrogen in the substrate resulted in significant increases in the nitrogen content of the foliage.

Increases in foliar nitrogen were correlated with increases in mite fecundity and developmental rate. Nitrogen supplied in amounts greater than that needed by the plant significantly stimulated mite fecundity and decreased the amount of time necessary for completion of the developmental period; an opposite effect occurred with deficient nitrogen supply.

Although DDT did not alter the foliar nitrogen content, it caused unknown physiological changes in

chrysanthemum foliage which affect the reproductive capacity of T. telarius.

SUMMARY

A review of the literature indicated that further investigation concerning a possible nitrogen-mite-DDT complex was warranted. Laboratory and greenhouse experiments were conducted to investigate the effects of DDT and nitrogen on chrysanthemums and, in turn, upon Tetranychus telarius (Linnaeus).

Plant response in vermiculite, sand, and soil was ascertained. Mite investigations were conducted on detached and isolated, intact foliage of plants grown in sand culture, and on naturally and artificially infested plants grown in soil under conditions which approached normal commercial greenhouse operations.

Nitrogen was applied to artificial cultures at 25-400 ppm. and to soil cultures at 0-400 ppm. In sand culture DDT was applied to the substrate at 0-40 pounds per acre; in soil studies DDT was applied to the substrate at 0 and 20 pounds per acre, and to the foliage as a 0.1 per cent (wettable) spray.

Mite developmental rate and fecundity revealed a highly significant positive response to nitrogen application and foliar nitrogen content.

DDT did not alter the nitrogen content of the foliage, but when applied to the substrate in sand cultures

did cause unknown physiological changes in the plants which affected the biotic potential of the mites. These effects were apparent at normal and surplus nitrogen levels, but not at deficient levels.

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APPENDIX

Tables 5 through 19

Analyses of variance have been completed by standard statistical procedures (Snedecor, 1956; Cochran and Cox, 1957). Each treatment group with significant differences is indicated by asterisks (* at the 95 per cent level of confidence and ** at the 99 per cent level of confidence). In addition, the least significant differences (LSD) at the 95 (.05) and 99 (.01) per cent levels of confidence are indicated when applicable. Finally, each treatment group which has a significant linear response is accompanied by a "b" value (regression coefficient) at the end of the analysis. The "b" value represents the average amount of change (positive or negative) found in the test organism per stated degree of treatment increase.

Table 5. Vermiculite series. Nitrogen content of foliage at five nitrogen levels (expressed as percentage of dry weight of foliage).

Group		PER CENT NITROGEN				
		Nitrogen in nutrient solution (ppm.)				
		25	50	100	200	400
I	Upper leaves	1.84	2.03	2.90	3.03	3.05
		1.93	1.96	2.52	2.91	3.06
		1.71	2.05	2.70	2.80	3.06
		1.67	1.89	2.49	2.67	3.12
	mean	1.79	1.98	2.65	2.85	3.07
	Lower leaves	1.35	1.62	2.80	2.88	3.01
		1.59	1.74	2.52	2.80	3.18
		1.35	1.78	2.56	2.68	3.18
		1.48	1.63	2.42	2.88	3.06
	mean	1.44	1.69	2.58	2.81	3.11
II		1.35	1.41	2.12	2.54	2.77
III		1.22	1.43	2.36	2.54	2.92
Apical meristems		2.48	2.45	2.70	2.98	3.04

Analysis of Variance
(Series I)

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Upper leaves					
Nitrogen	4	4.984	1.246**	0.19	0.26
linear response	1	3.735	3.735**		
deviation	3	1.249	0.416**		
Error	15	0.240	0.016		
Lower leaves					
Nitrogen	4	8.356	2.089**	0.17	0.23
linear response	1	6.241	6.241**		
deviation	3	2.115	0.705**		
Error	15	0.186	0.012		

b : Upper leaves $\neq$ 0.08% N/25 ppm. nitrogen
Lower leaves $\neq$ 0.10% N/25 ppm. nitrogen

Table 6. Nitrogen series. Number of offspring of three adult female mites at five nitrogen levels.

Nutrient method	NUMBER OF OFFSPRING Parts per million of nitrogen 25	50	100	200	400
A	39	5	8	63	5
	9	31	43	53	106
	10	11	23	53	86
	3	13	31	14	51
mean	15	15	26	46	62
B	23	11	49	89	70
	5	25	70	75	61
	24	38	55	47	140
	11	12	63	71	88
mean	16	22	59	70	90

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Method A					
Nitrogen	4	6753	1688		
linear response	1	6452	6452**		
deviation	3	301	100		
Error	15	9096	606		
Method B					
Nitrogen	4	16249	4062**	29	40
linear response	1	13319	13319**		
deviation	3	2930	977		
Error	15	5656	377		

b : Method A $\neq$ 3.29 mites/25 ppm. nitrogen
Method B $\neq$ 4.73 mites/25 ppm. nitrogen

Table 7. Nitrogen series. Length of developmental period (egg to adult female mite) at five nitrogen levels.

Nutrient method	TIME IN HOURS Parts per million of nitrogen 25	50	100	200	400
A	501	501	348	288	288
	428	348	348	288	255
	-	-	307	254	288
	-	-	-	288	255
mean	464	424	334	280	272
B	287	287	-	190	211
	542	380	350	237	237
	-	288	238	287	207
	-	-	399	212	238
mean	414	329	318	232	223

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Method A					
Nitrogen	4	77905	19476**	77	109
linear response	1	64236	64236**		
deviation	3	13669	4556		
Error	10	17446	1745		
Method B					
Nitrogen	4	69326	17332		
linear response	1	56671	56671**		
deviations	3	12655	4218		
Error	11	57874	5261		

b : Method A - 11.9 hours/25 ppm. nitrogen
Method B - 11.0 hours/25 ppm. nitrogen

Table 8. Nitrogen x DDT (detached) series. Number of offspring of three adult female mites at four nitrogen levels and six DDT levels.

Nitrogen (ppm.)	NUMBER OF OFFSPRING						
	Pounds of DDT per acre						
	0	0.9	5	10	20	40	mean
50	24	32	14	45	41	57	
	32	47	57	35	28	41	
	29	46	49	41	31	37	38
	49	29	51	34	27	34	
100	69	30	15	42	24	50	
	93	33	25	52	39	107	
	53	57	19	36	66	59	50
	37	55	44	58	100	42	
200	74	91	51	55	81	103	
	107	82	51	41	106	96	
	74	114	79	67	93	73	80
	86	87	61	104	80	77	
400	203	137	61	56	51	82	
	71	97	97	100	109	101	
	115	115†	70	103	103	142	105
	110	98	125	85	141	141	
mean	77	69	54	60	70	78	

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Replication	3	1556	519		
Nitrogen	3	65421	21807**	13	18
linear response	1	62220	62220**		
deviation	2	3201	1600		
DDT	5	7086	1417*	16	22
Nitrogen x DDT	15	6153	410		
Error	68	35950	529		

b : Nitrogen $\hat{}$ 9.5 mites/50 ppm. nitrogen

† Missing datum, calculated.

Table 9. Nitrogen x DDT (detached) series. Length of developmental period (egg to adult female mite) at four nitrogen levels and six DDT levels.

Nitrogen (ppm.)	TIME IN HOURS						
	Pounds of DDT per acre						
	0	0.9	5	10	20	40	mean
50	398	494	-	-	256	515	
	334	403	403	256	309	309	
	-	346	-	423,283'	399	327	364
	323	397	-	376	-	-	
100	303	-	-	346	-	372	
	210	425	333	333	403	280	
	378	327	301	301	301	378	335
	255	352	229	281	352	568	
200	281	326	-	256	281	327	
	210	256	309	309	233	233	
	252	253	283	280	252	252	275
	255	324	323	323	255	255	
400	232	232	302	256	232	206	
	256	256	233	210	211	233	
	253	-	252	252	252	234	246
	230	229	255	255	301	280	
mean	278	330	293	296	288	318	

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Replication	3	5877	1959		
Nitrogen	3	180739	60246**	33	44
linear response	1	165460	165460**		
deviation	2	15279	7640		
DDT	5	27305	5461		
Nitrogen x DDT	15	42197	2813		
Error	58	171418	2955		

b : Nitrogen - 16.4 hours/50 ppm. nitrogen

' Two plates, both included in analysis.

Table 10. Nitrogen x DDT (detached) series. Length of developmental period (egg to egg) at four nitrogen levels and six DDT levels.

Nitrogen (ppm.)	TIME IN HOURS Pounds of DDT per acre 0	0.9	5	10	20	40	mean
50	425	-	-	-	446	-	
	447	-	-	280	334	334	
	-	497	-	328	471	378	408
	422	496	-	448	-	-	
100	446	-	-	493	-	473	
	233	517	403	381	517	334	
	423	378	-	-	347	-	398
	301	-	281	352	377	-	
200	303	-	-	281	302	446	
	233	308	333	334	280	257	
	300	280	328	301	280	327	301
	255	352	352	-	280	301	
400	281	281	347	281	256	233	
	308	309	280	257	233	280	
	280	-	280	280	280	252	287
	352	352	301	301	-	352	
mean	334	377	323	332	339	331	

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Replication	3	9139	3046		
Nitrogen	3	185018	61673**	43	57
linear response	1	151581	151581**		
deviation	2	33437	16718*		
DDT'	4	16110	4028		
Nitrogen x DDT	12	46705	3892		
Error	40	139148	3479		

b : Nitrogen - 18.2 hours/50 ppm. nitrogen

' 5 pounds DDT/acre data not included.

Table 11. Nitrogen x DDT (detached) series. Length of developmental period (egg to larva of succeeding generation) at four nitrogen levels and six DDT levels.

	TIME IN HOURS						
Nitrogen (ppm.)	Pounds of DDT per acre 0	0.9	5	10	20	40	mean
50	545	-	-	-	545	-	
	551	-	-	381	425	403	
	-	588	-	423	563	447	500
	495	590	-	543	-	-	
100	568	-	-	568	-	545	
	309	593	497	469	-	425	
	515	471	-	-	447	-	481
	422	-	352	448	473	-	
200	398	-	-	372	398	545	
	334	381	403	403	357	357	
	378	379	400	378	346	447	392
	352	473	448	-	377	376	
400	371	372	446	372	303	303	
	381	381	357	334	334	357	
	347	-	346	346	346	346	361
	423	448	377	377	-	-	
mean	426	468	403	416	410	414	

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Replication	3	22933	7644		
Nitrogen	3	202971	67597**	40	53
linear response	1	180699	180699**		
deviation	2	22092	11046*		
DDT'	4	23384	5846		
Nitrogen x DDT	12	48145	4012		
Error	38	109660	2886		

b : Nitrogen - 20.3 hours/50 ppm. nitrogen

' 5 pounds DDT/acre data not included.

Table 12. Nitrogen x DDT (intact) series. Nitrogen content of foliage at five nitrogen levels (expressed as percentage of dry weight of foliage).

	PER CENT NITROGEN				
	Nitrogen in nutrient solution (ppm.)				
	25	50	100	200	400
	1.60	1.37	2.56	3.69	3.28
	1.35	1.38	2.28	3.29	4.22
	1.32	1.48	2.39	3.84	3.39
	1.16	1.40	2.67	3.88	3.22
mean	1.36	1.41	2.48	3.68	3.52

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Replication	3	0.1	0.03		
Nitrogen	4	19.8	4.95**	0.44	0.66
linear response	1	13.9	13.94**		
deviation	3	5.9	1.97**		
Error	12	1.0	0.08		

b : ≠ 0.15% N/25 ppm. nitrogen

Table 13. Nitrogen x DDT (intact) series. Number of offspring at five nitrogen levels.

Period in days	NUMBER OF OFFSPRING				
	Nitrogen in nutrient solution (ppm.)				
	25	50	100	200	400
14	43	87	45	113	104
	15	50	84	87	94
	4	21	45	136	54
	51	59	54	110	75
mean	28	54	57	112	82
27	25	136	335	721	248
	3	62	448	621	373
	15	167	122	304	667
	38	161	354	542	484
mean	20	132	315	547	443

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
14 day period					
Replication	3	1815	605		
Nitrogen	4	15844	3961**	33	47
linear response	1	6766	6766**		
deviation	3	9078	3026**		
Error	12	5594	466		
27 day period					
Replication	3	10114	3371		
Nitrogen	4	751798	187950**	220	308
linear response	1	424951	424951**		
deviation	3	236847	108949*		
Error	12	243996	20333		

b : 14 day period $\neq$ 3.4 mites/25 ppm. nitrogen
27 day period $\neq$ 26.7 mites/25 ppm. nitrogen

Table 14. Nitrogen x DDT (intact) series. Ranked stage of development after 14 days at five levels of nitrogen.

	STAGE OF DEVELOPMENT (RANKED)'				
	Nitrogen in nutrient solution (ppm.)				
	25	50	100	200	400
	1	3	3	5	5
	1	3	5	4	4
	1	2	4	4	4
	2	3	4	5	5
mean	1.2	2.8	4.0	4.5	4.5

Analysis of Variance

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Replication	3	1.6	.53		
Nitrogen	4	31.3	7.82**	0.9	1.2
linear response	1	17.3	17.3 **		
deviation	3	14.0	4.67**		
Error	12	3.9	0.32		

b : $\pm$ 0.17 ranks/25 ppm. nitrogen

' Key for ranking:

Rank	Developmental stage
0	Egg
1	Larva
2	quiescent Larva
3	Protonymph
4	quiescent Protonymph
5	Deutonymph
6	quiescent Deutonymph
7	Adult (female)

Table 15. Nitrogen x DDT (intact) series. Results of one replication at four nitrogen levels and five DDT levels.

A. Number of offspring of three adult female mites after 14 (in brackets) and 27 days.

	NUMBER OF OFFSPRING			
DDT	Nitrogen in nutrient solution (ppm.)			
lbs./A.	50	100	200	400
0.9	27 (77)	591 (110)	736 (87)	465 (43)
5	6 (28)	270 (75)	772 (88)	871 (99)
10	337 (36)	536 (92)	733 (120)	713 (131)
20	119 (26)	520 (85)	695 (60)	293 (113)
40	70 (45)	256 (46)	198 (94)	590 (82)
mean	112 (42)	433 (82)	627 (90)	586 (94)

B. Nitrogen content of foliage (expressed as percentage of dry weight of foliage).

	PER CENT NITROGEN			
DDT	Nitrogen in nutrient solution (ppm.)			
lbs./A.	50	100	200	400
0.9	1.63	1.65	2.90	4.04
5	2.43	2.57	2.92	3.84
10	1.56	2.29	3.66	3.93
20	1.33	2.39	3.04	4.01
40	1.55	2.26	3.83	3.43
mean	1.70	2.23	3.27	3.85

C. Ranked stage of development after 14 days.*

	STAGE OF DEVELOPMENT (RANKED)*			
DDT	Nitrogen in nutrient solution (ppm.)			
lbs./A.	50	100	200	400
0.9	3	4	5	4
5	3	4	5	7
10	2	5	5	7
20	1	3	5	7
40	2	3	7	4
mean	2.2	3.8	5.4	5.8

* See key for ranking, Table 14.

Table 16. Liquid Nitrogen series. Nitrogen content of foliage at four nitrogen levels and three DDT levels (expressed as percentage of dry weight of foliage).

DDT treatment	PER CENT NITROGEN Liquid nitrogen applied (ppm.) 0	50	200	400
None	2.36	2.77	3.19	3.18
	2.30	2.44	3.03	3.55
	2.47	2.51	2.91	3.33
	2.43	2.66	3.11	3.30
mean	2.39	2.60	3.06	3.34
Spray		2.74	2.93	3.60
		2.52	3.08	3.76
		2.36	2.96	3.28
		2.44	3.25	3.16
mean		2.52	3.06	3.45
Soil		2.70	3.06	3.30
		2.44	3.03	3.21
		2.45	3.00	3.41
		2.45	2.82	3.38
mean		2.51	2.98	3.32

Analysis of Variance '

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Plots					
Blocks	3	0.10	0.033		
Nitrogen	2	4.20	2.100**	0.17	0.25
linear response	1	4.05	4.05 **		
deviation	1	0.15	0.15		
Error (a)	6	0.17	0.028		
Subplots					
DDT	2	0.04	0.020		
Nitrogen x DDT	4	0.03	0.008		
Error (b)	18	0.39	0.022		

b : Nitrogen / 0.12% N/50 ppm. nitrogen

' Split plot design, 0 ppm. nitrogen level omitted.

Table 17. Liquid Nitrogen series. Mean number of mites of two replicates per plot at four nitrogen levels and three DDT treatments.

DDT treatment	MEAN NUMBER OF MITES Liquid nitrogen applied (ppm.) 0	50	200	400
None	1008	4036	5076	6600
	1802	1988	4140	7580
	796	2120	8824	6532
	2388	4120	8424	9404
mean	1498	3066	6616	7529
Spray		3264	6624	8308
		3664	7156	6556
		2024	14240	6928
		3688	10408	7320
mean		3160	9607	7278
Soil		4156	1132	7352
		2964	5264	5664
		2332	4920	10064
		4668	7820	10816
mean		3530	4784	8474

Analysis of Variance '

Source of Variation	Degrees Freedom	Sum of Squares	Mean Square	LSD .05	LSD .01
Plots					
Blocks	3	1372	457		
Nitrogen	2	6517	3258*		
50 vs 200, 400	1	6257	6257**		
200 vs 400	1	260	260		
linear response	1	5505	5505**		
deviation	1	1012	1012		
Error (a)	6	1840	307		
Subplots					
DDT	2	337	169		
Nitrogen x DDT	4	1666	416**		
DDT x 50 vs DDT x 200, 400	1	316	316		
DDT x 200 vs DDT x 400	1	1350	1350**		
Error (b)	18	1494	83		

' Split plot design, 0 ppm. nitrogen level omitted; all data transformed into square roots for analysis.

Table 18. Liquid Nitrogen series. Mean results of soil analyses.

DATE	TREATMENT N (ppm.)	TREATMENT DDT	PARTS PER MILLION NO_3	NH_3	P	K	Ca	pH	SS
Jun 14	0-400	1,2,3*	35	12	88	75	1500	5.4	53
Jul 1	0	1	10	12	150	30	1600	5.7	59
	200	1	20	12	150	60	1600	5.7	58
	200	2	10	12	100	60	1600	5.7	44
	400	3	10	12	100	30	1600	5.7	58
Jul 19	0	1	16	16	75	140	1600	6.0	32
	50	1,2	12	17	75	68	1825	6.0	28
	200	1,2	51	54	62	195	1600	5.8	55
	400	1,2	51	169	75	168	1600	6.0	38
Aug 5	0	1	4	6	75	180	1150	6.2	20
	50	1,2	20	6	44	120	1325	5.8	48
	200	1,2	21	42	69	138	1425	5.8	35
	400	1,2	39	8	38	82	1175	5.6	47
Sep 16	0	1	15	8	56	300	1625	6.1	38
	50	1,2,3	14	6	50	206	1350	5.9	28
	200	1,2,3	82	8	65	153	1600	5.2	37
	400	1,2,3	200	16	65	144	1708	4.7	87

* Key for DDT treatments:

1. No DDT applied.
2. DDT applied to soil.
3. DDT applied as spray.

Table 19. Foliar content of potassium, phosphorus, calcium, and magnesium (expressed as percentage of dry weight of foliage).

Series	TREATMENT N (ppm.)	TREATMENT DDT	PER CENT OF DRY WEIGHT Ca	K	P	Mg
Vermiculite	25	1*	0.9	4.1	0.27	0.30
	50	1	0.8	4.0	0.30	0.35
	100	1	0.9	4.4	0.30	0.44
	200	1	1.0	4.6	0.33	0.51
	400	1	1.0	5.0	0.26	0.58
Nitrogen Method A & B combined	25	1	0.8	4.0	0.27	0.19
	50	1	0.9	4.2	0.33	0.25
Method A	100	1	1.0	4.3	0.31	0.23
	200	1	0.7	4.8	0.27	0.39
	400	1	1.4	5.2	0.24	0.28
Method B	100	1	1.2	4.7	0.28	0.25
	200	1	0.7	4.4	0.27	0.44
	400	1	0.8	4.7	0.28	0.29
Liquid Nitrogen	0	1	1.4	4.1	0.23	0.58
	50	1	1.2	4.8	0.22	0.58
	50	2	1.3	5.0	0.24	0.49
	50	3	1.2	5.2	0.24	0.29
	200	1	1.3	5.2	0.27	0.54
	200	2	1.3	5.5	0.27	0.44
	200	3	1.3	5.6	0.28	0.47
	400	1	1.2	5.4	0.29	0.50
	400	2	1.3	5.5	0.30	0.47
	400	3	1.3	5.6	0.30	0.43

* Key for DDT treatments:

1. No DDT applied.
2. DDT applied to soil.
3. DDT applied as spray.

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